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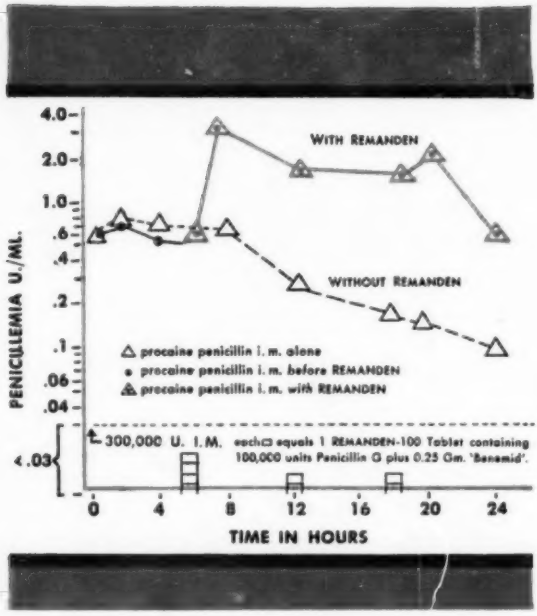

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THE SENSITIVITY OF PATHOGENIC BACTERIA TO A SERIES OF ANTIBIOTICS[1]

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The increasing use of antibiotics in clinical medicine and the development of new antimicrobial drugs have brought into sharp focus the problem of bacterial resistance to these therapeutic agents (1). This difficulty arises most commonly in infections caused by staphylococci or Gram negative bacilli. Disregarding several important factors such as the origin of resistant strains, epidemiology of nosocomial infection, the role of surgical drainage, and so-called "host factors" in resistance, it can be said that the therapeutic enigma presented by resistant bacterial infections has led to emphasis upon two developments:

(1) A tremendous increase in the use of laboratory tests as aids in the selection of antibiotics for clinical use.

(2) Continuing evaluation in the laboratory and clinic of newly developed drugs that show some promise of activity against organisms that resist the agents presently in use.

The use of the "disk" method or its variants on a routine basis provides a reliable guide in most situations, although this procedure has certain shortcomings (2). Disks are often impregnated with drug in a concentration lower than that obtainable *in vivo* by large dosage or parenteral routes and, particularly in the case of penicillin, it may be concluded erroneously that the drug will be clinically ineffective. Furthermore, depending as it does upon diffusion of the drug through agar, the disk method can be misleading with substances such as bacitracin, neomycin, and polymyxin, in the presence of which even highly sensitive strains often show relatively narrow circles of inhibition. There is little opportunity to distinguish between bacteriostatic and bactericidal concentrations of an agent, an important therapeutic consideration in several types of serious infection. Finally, in order to compare the efficacy of several agents quantitatively, serial dilutions in liquid media and a specific end-point are highly desirable. This applies particularly to any comparison of newly available agents with the drugs of known efficacy.

This report details the results of *in vitro* tests of 100 microörganisms against a variety of antibiotic agents, employing a tube-dilution test and determining both bacteriostatic and bactericidal end-points. All strains of bacteria tested

[1] Supported by a contract with the Army Chemical Corps, Fort Detrick, Maryland.

[2] Research Fellow of the National Foundation for Infantile Paralysis.

TABLE I

Sensitivity Studies on 68 Strains of Staphylococcus

#	Organism	Pen. u/ml.	Tet. μg/ml.	Chl. μg/ml.	Ery. μg/ml.	Bac. u/ml.	Neo. μg/ml.	Spir. μg/ml.	Ami. μg/ml.	Cath. μg/ml.	Alba. μg/ml.
1	Hem. staph. aureus	30/70	50/N	10/N	1/50	1/1	5/5	20/20	5/N	1/1	.2/50
2	Hem. staph. aureus	10/30	100/N	10/N	20/100	1/10	5/20	100/N	50/N	.2/.2	.2/.2
3	Hem. staph. aureus	5/70	100/100	10/N	1/50	5/50	5/10	20/N	5/50	1/1	.2/10
4	Hem. staph. aureus	5/N	1/50	10/50	1/20	50/50	10/10	50/N	5/100	.2/100	1/100
5	Hem. staph. aureus	250/N	>100	20/100	>100	5/20	1/5	5/N	5/N	.2/50	.2/50
6	Hem. staph. aureus	5/N	>100	20/N	1/20	5/10	1/100	10/N	5/N	1/20	1/100
7	Hem. staph. aureus	100/N	100/100	50/N	1/100	20/50	50/100	50/N	5/N	1/100	5/N
8	Hem. staph. aureus	70/250	1/10	10/50	20/20	10/N	20/N	20/N	1/N	1/N	.2/50
9	Hem. staph. aureus	250/N	>100	20/N	20/N	5/5	5/5	100/N	>100	5/100	1/100
10	Hem. staph. aureus	>250	100/N	20/N	1/N	5/20	50/100	20/N	5/N	1/N	5/100
11	Hem. staph. aureus	1/40	5/50	20/100	5/50	5/100	5/5	>100	100/N	1/20	.2/20
12	Hem. staph. aureus	250/N	>100	10/50	1/100	10/50	5/20	10/N	5/100	1/100	1/N
13	Hem. staph. aureus	20/90	1/50	10/N	1/20	10/10	10/10	50/50	5/100	.2/100	.2/100
14	Hem. staph. aureus	5/100	1/100	10/100	1/100	10/20	5/50	10/N	5/N	.2/100	.2/50
15	Hem. staph. aureus	50/250	100/100	10/20	20/100	5/10	5/10	50/N	50/N	1/10	.2/50
16	Hem. staph. aureus	5/90	100/100	10/100	>100	5/100	10/10	20/N	5/5	1/10	.2/20
17	Hem. staph. aureus	5/30	>100	10/N	>100	10/20	20/20	20/20	5/50	.2/N	.2/10
18	Hem. staph. aureus	60/N	5/50	10/100	1/20	10/100	5/5	20/N	5/10	1/20	.2/10
19	Hem. staph. aureus	20/100	>100	10/N	>100	10/10	5/20	50/N	10/N	.2/20	.2/N
20	Hem. staph. aureus	50/N	>100	10/20	10/N	10/10	5/10	10/N	5/100	.2/N	1/N
21	Hem. staph. aureus	5/250	1/N	10/N	1/N	20/20	10/10	50/50	5/N	.2/N	1/50
22	Hem. staph. aureus	.5/60	1/5	10/100	1/N	50/50	20/20	20/N	5/100	.2/N	1/100
23	Hem. staph. aureus	5/50	50/100	10/100	5/50	10/100	10/10	20/N	5/N	.2/100	.2/50
24	Hem. staph. aureus	250/N	100/N	10/N	5/N	10/20	10/20	50/N	5/N	.2/N	1/N
25	Hem. staph. aureus	250/N	100/100	10/N	.2/N	5/10	10/10	10/N	5/N	.2/50	.2/N
26	Hem. staph. aureus	80/250	100/100	10/100	20/N	5/10	10/20	100/N	50/N	.2/N	1/50
27	Hem. staph. aureus	5/70	1/N	10/100	1/20	20/100	10/10	20/N	5/50	.2/10	1/20
28	Hem. staph. aureus	20/250	50/100	10/N	1/N	1/1	5/20	50/N	10/N	.2/10	.2/10
29	Hem. staph. aureus	20/100	50/N	10/N	10/N	.2/10	5/5	>100	100/N	.2/1	.2/1
30	Hem. staph. aureus	5/5	1/20	10/N	1/10	5/10	5/5	10/N	5/N	1/100	1/100
31	Hem. staph. aureus	20/N	>100	10/N	.2/10	1/100	50/100	5/N	1/100	20/50	20/100
32	Hem. staph. aureus	10/N	1/5	10/100	1/1	5/10	5/50	10/50	5/20	.2/N	.2/N
33	Hem. staph. aureus	90/N	50/N	5/N	.2/2	1/5	5/5	10/100	1/100	.2/20	.2/5
34	Hem. staph. aureus	.5/20	1/5	10/20	.2/5	10/10	10/20	10/N	1/20	1/100	1/1
35	Hem. staph. aureus	1/5	50/50	5/100	.2/1	5/10	1/5	10/N	1/10	1/1	1/1
36	Hem. staph. aureus	30/N	50/N	10/N	1/20	1/50	5/50	10/N	1/N	1/100	1/100
37	Hem. staph. aureus	30/N	50/100	5/N	.2/50	1/5	5/5	10/N	1/N	1/50	1/100
38	Hem. staph. aureus	20/50	>100	5/50	1/1	1/10	1/5	10/100	1/1	.2/20	.2/1

34	Hem. staph. aureus	.5/20	1/5	10/20	.2/5	10/10	10/20	10/N	1/20	1/100	1/1
35	Hem. staph. aureus	1/5	50/50	5/100	.2/1	5/10	1/5	10/N	1/10	1/1	1/1
36	Hem. staph. aureus	30/N	50/N	10/N	1/20	1/50	5/50	10/N	1/N	1/100	1/100
37	Hem. staph. aureus	30/N	50/100	5/N	.2/50	1/5	5/5	10/N	1/N	1/50	1/100
38	Hem. staph. aureus	20/50	>100	5/50	1/1	1/10	1/5	10/100	1/1	.2/20	.2/1
39	Hem. staph. aureus	30/50	50/100	5/N	>100	5/5	20/20	10/N	5/N	.2/50	.2/50
40	Hem. staph. aureus	30/70	100/100	5/N	>100	5/5	20/N	20/N	5/N	.2/100	.2/100
41	Hem. staph. aureus	20/50	100/100	5/100	>100	5/10	50/50	20/N	5/N	1/100	1/50
42	Hem. staph. aureus	250/N	100/100	5/100	1/100	5/5	1/5	10/N	1/N	1/100	1/100
43	Hem. staph. aureus	40/N	>100	5/N	>100	5/10	5/10	10/N	5/N	.2/100	1/N
44	Hem. staph. albus	10/70	>100	50/N	>100	5/10	5/20	10/N	>100	1/N	1/50
45	Hem. staph. albus	10/30	>100	5/50	>100	5/20	5/50	20/N	>100	1/10	.2/20
46	Hem. staph. albus	20/30	>100	5/100	>100	5/10	5/20	10/N	>100	.2/1	.2/.2
47	Hem. staph. albus	—	—	—	—	—	—	—	—	—	.2/50
48	Hem. staph. albus	20/250	1/20	5/N	1/50	1/20	1/10	10/N	1/N	5/10	1/50
49	Hem. staph. albus	30/250	1/20	50/N	1/20	5/50	1/5	10/N	1/N	1/50	.2/20
50	Hem. staph. albus	30/N	>100	5/N	1/50	5/10	1/5	10/N	1/20	1/N	1/N
51	Hem. staph. albus	10/70	1/10	5/100	1/50	5/50	5/50	10/100	1/N	1/N	.2/100
52	Hem. staph. albus	—	—	—	—	—	—	—	—	—	1/N
53	Hem. staph. albus	10/80	50/N	5/N	>100	5/N	5/20	20/N	5/N	1/100	1/N
54	Hem. staph. albus	1/5	50/50	10/10	1/1	5/5	1/1	5/10	1/1	1/1	.2/.2
55	Hem. staph. albus	.5/20	1/50	10/50	1/50	5/100	5/20	5/N	5/50	1/100	.2/50
56	Hem. staph. albus	10/30	>100	10/100	>100	10/20	5/20	50/N	>100	.2/N	.2/5
57	Hem. staph. albus	—	—	—	1/50	—	—	—	—	—	1/20
58	Hem. staph. albus	90/N	1/100	10/N	.2/50	5/100	1/10	5/N	1/100	1/100	1/20
59	Hem. staph. albus	—	—	—	—	—	—	—	—	—	.2/.2
60	Non-hem. staph. aureus	1/5	>100	10/10	>100	10/10	50/50	50/50	5/5	1/1	.2/.2
61	Non-hem. staph. aureus	5/60	100/N	10/N	1/50	10/50	5/5	20/100	5/N	1/100	.2/100
62	Non-hem. staph. aureus	5/250	>100	10/N	1/50	10/100	10/50	10/N	5/100	.2/10	.2/N
63	Non-hem. staph. aureus	5/70	1/10	5/100	1/50	5/10	20/20	50/N	5/N	.2/10	1/10
64	Non-hem. staph. aureus	5/250	5/10	10/N	.2/20	5/N	20/50	10/N	5/N	1/50	1/50
65	Non-hem. staph. aureus	5/90	1/N	10/N	.2/10	5/N	5/10	10/N	1/N	1/N	1/100
66	Non-hem. staph. aureus	—	—	—	—	—	—	—	—	—	1/5
67	Non-hem. staph. albus	10/10	100/100	5/5	>100	1/5	1/1	10/50	5/10	1/5	1/10
68	Non-hem. staph. albus	.2/.5	1/50	5/N	1/20	1/5	.2/100	10/20	1/100	1/20	1/1

Numerator: Minimal inhibitory (bacteriostatic) concentration.
Denominator: Bactericidal concentration.
N: No bactericidal effect.
—: Not done.

were isolated from severe infections that presented therapeutic problems of more than average difficulty. The correlation of these data with the patients' clinical course and therapeutic response and the relationship of antibiotic therapy to the development of resistance will be covered in a separate report.

MATERIALS AND METHODS

One hundred strains of bacteria isolated from 78 patients were tested. Eighty were Gram positive, including 68 staphylococci (Table I) and 12 streptococci and others (Table II). The staphylococci came from the following sources: blood, 12; sputum, 18; abscesses, 27; urine, 7; other purulent foci, 4. The remaining Gram positive organisms were isolated as follows: blood, 5; urine, 5 (all enterococci); sputum 1, and brain abscess, 1. There were 20 Gram negative bacilli as listed in Table III which were cultured from the following sites: blood, 8; urine, 9; sputum, 1; abscesses, 2.

Gram positive organisms, with occasional exceptions, were tested against penicillin, erythromycin, tetracycline, chloramphenicol, bacitracin, neomycin, spiramycin (Merck), Amimycin (oleandomycin, Pfizer), Cathomycin (novobiocin, Merck), and Albamycin (novobiocin, Upjohn). The usual drugs for assaying Gram negative bacilli were neomycin, polymyxin, streptomycin, tetracycline, chloramphenicol, novobiocin, and, occasionally, penicillin (see Table III).

The organism to be assayed was grown in trypticase soy broth at 37°C. for 18 hours and an aliquot of this culture was diluted 1:1000 in fresh trypticase soy broth immediately before tests were performed. This diluted inoculum was added in the amount of 0.1 ml. to each of a series of Kahn tubes containing 0.8 ml. of broth and 0.1 ml. of a stock dilution of an antibiotic. The final volume in the test system was therefore 1.0 ml., and each drug except penicillin was employed in final concentrations of 0.2, 1.0, 5.0, 10, 20, 50 and 100 mcgm. or units/ml. The concentrations of penicillin were 0.2, 0.5, 1.0, 5.0, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, and 250 units/ml. After incubation at 37°C. for 24 hours, the tubes were examined for evidence of growth. The first tube showing no turbidity was taken as the bacteriostatic end-point. A single loopful of material from all tubes that remained clear was inoculated onto blood agar; the concentration at which no colonies grew on the agar after incubation for 24 hours at 37°C. was taken as the bactericidal end-point. Stock dilutions of each antibiotic were prepared in quantity and stored at −20°C. In many tests, the entire series of tubes containing broth and drugs had been prepared ahead of time and frozen at −20°C. until used.

RESULTS

The pertinent information for the 100 microörganisms tested is shown in Table I–III. The bacteriostatic concentration of each drug is the numerator; the denominator indicates the bactericidal level. When killing was incomplete, the denominator is designated "N".

Staphylococci.—The predominance of these organisms, of which 68 strains were tested, emphasizes the current importance of the staphylococcus in the etiology of resistant bacterial infections. No sub-division into albus or aureus varieties has been attempted in this analysis because all were clearly responsible for severe infections. The high degree of resistance to penicillin of most of these strains is summarized in Table IV. More than 30 per cent of the strains sur-

TABLE II

Sensitivity of Gram Positive Organisms Other Than Staphylococci

#	Organism	Pen.	Tet.	Chl.	Ery.	Bac.	Neo.	Spir.	Ami.	Cath.	Alba.
69	Alpha streptococcus	.2/.2	.2/1	.2/1	.2/.2	.2/.2	.2/.2	.2/.2	.2/.2	1/10	1/10
70	Gram positive rod (? Listeria)	1/1	5/N	5/N	.2/100	1/N	10/50	1/N	.2/N	>100	>100
71	Alpha streptococcus	5/5	1/1	10/10	.2/.2	5/5	50/50	5/5	1/1	1/1	1/1
72	Beta hemolytic streptococcus	.2/.2	.2/5	.2/.2	.2/.2	.2/.2	.2/5	.2/.2	.2/1	.2/10	5/20
73	Alpha streptococcus	10/10	1/1	1/5	.2/.2	1/5	5/5	.2/.2	.2/.2	1/5	1/5
74	Streptococcus fecalis	20/30	1/N	5/N	1/50	.2/.2	20/20	5/N	5/N	10/N	20/100
75	Streptococcus fecalis	—	—	—	—	—	—	5/N	—	—	20/N
76	Streptococcus fecalis	—	—	—	—	—	—	5/N	—	—	20/N
77	Streptococcus fecalis	—	—	—	—	—	—	—	—	—	20/N
78	Streptococcus fecalis	—	—	—	—	—	—	—	—	—	>100
79	Streptococcus fecalis	—	—	—	—	—	—	—	—	—	10/N
80	Streptococcus fecalis	—	—	—	—	—	—	1/N	—	—	20/N

TABLE III

Sensitivity Studies on Gram Negative Bacteria

#	Organism	Pen.	Tet.	Chl.	Ery.	Bac.	Neo.	Spir.	Ami.	Cath.	Alba.	Poly.	Strep.
81	E. coli	—	—	—	—	—	—	—	—	—	>100	—	—
82	Paracolon	>250	>100	50/N	100/N	>100	50/50	>100	—	>100	>100	—	—
83	E. coli	—	—	—	—	—	—	—	—	—	100/N	—	—
84	Paracolon	—	—	—	—	—	—	—	—	—	100/N	—	—
85	E. coli	—	—	—	—	—	—	—	—	—	>100	—	—
86	Paracolon	>250	50/N	50/N	—	—	>100	—	—	—	>100	20/50	>100
87	Paracolon	70/70	5/100	5/N	—	—	100/100	—	—	—	>100	20/20	100/N
88	E. coli	—	5/N	>100	—	—	50/100	—	—	—	>100	50/50	>100
89	S. cholerasuis	30/30	10/100	5/N	—	—	100/100	—	—	—	>100	10/20	>100
90	Alkaligenes Fecalis	—	20/50	>100	—	—	20/50	—	—	—	>100	20/50	>100
91	Alkaligenes Fecalis	—	—	—	—	—	—	—	—	—	>100	—	—
92	Alkaligenes Fecalis	—	10/50	>100	—	—	50/100	—	—	>100	—	50/50	50/100
93	Alkaligenes Fecalis	—	10/50	100/N	—	—	50/50	—	—	>100	—	50/50	50/100
94	Alkaligenes Fecalis	—	5/5	20/50	—	—	>100	—	—	>100	—	50/50	100/N
95	Proteus	—	—	—	—	—	—	—	—	—	>100	—	—
96	Proteus	—	—	—	—	—	—	—	—	—	>100	—	—
97	Proteus	—	50/50	10/10	—	—	—	—	—	—	10/10	—	>100
98	Pseudomonas	>250	50/100	100/N	>100	>100	50/N	>100	>100	>100	>100	20/20	—
99	Pseudomonas	—	20/50	>100	—	—	50/N	—	—	—	—	20/20	>100
100	Pseudomonas	—	20/50	>100	—	—	50/50	—	—	—	>100	20/50	100/100

TABLE IV

Sensitivity of 68 Strains of Staphylococci to Penicillin

Concentration u./ml.	.2	.5	1	5	10	20	30	40	50	60	70	80	90	100	250	None
No. Strains Killed	0	1	0	3	3	3	4	1	4	3	6	0	4	3	9	19
Cumulative Per Cent Killed	0	1.6	1.6	6.3	11.1	15.9	22.1	23.9	30.2	35.0	44.4	44.4	50.7	55.6	69.8	—
No. Strains Inhibited	1	3	2	16	9	7	7	1	3	1	2	1	2	1	5	2
Cumulative Per Cent Inhibited	1.6	6.3	9.5	35.0	49.3	60.3	71.5	73.0	77.8	79.4	82.6	84.2	87.3	89.0	96.8	—

vived a concentration of 250 units/ml. of penicillin and only 30.4 per cent were *killed* by 50 units/ml. or less. If one considers failure to produce turbidity in broth after 24 hours, the *bacteriostatic* end-point, the situation appears to be better in that 78 per cent of the strains were inhibited by 50 units/ml. and all but 2 by 250 units/ml. However, the importance of direct *killing* action of antibiotics in severe staphylococcal infections is such that the bactericidal level is probably a better indicator of the effectiveness of an antimicrobial drug against this organism.

Bacitracin and neomycin were by far the most potent *bactericidal* agents tested although several others were as efficacious as *inhibitors* of growth.

Tetracycline was *bacteriostatic* in low concentrations against about one-third of the strains, the remainder requiring very high concentrations for any effect at all. Chloramphenicol, in the range of 5.0–10 mcgm./ml. was an excellent *inhibitor* of growth for the majority of staphylococci tested. Tetracycline and chloramphenicol were relatively impotent *killing* agents even in concentrations of 100 mcgm./ml.

Erythromycin was effective in producing *bacteriostasis* in 70 per cent of the tests but 15 strains proved to be completely resistant to the concentrations employed.

Of particular interest were the results with spiramycin, Amimycin (oleandomycin), and the two novobiocin preparations, Cathomycin and Albamycin. High concentrations of spiramycin were needed for *inhibition*, and *killing* was rarely seen. Amimycin is best described as similar in its range of activity to erythromycin in this series of tests. Novobiocin was strikingly effective in its *bacteriostatic* action, all but one strain being inhibited by 5 mcgm./ml. and all but 4 by 1.0 mcgm./ml. Although not as uniformly effective as bacitracin and neomycin, novobiocin was, nonetheless, impressively *bactericidal* against many strains. The single strain that showed relative resistance to this drug (Table I, No. 31) was isolated from the sputum of a patient with pneumonia who had never been given novobiocin.

The sensitivity patterns for the several antibiotics tested are shown graphically in Figures 1 and 2, and are summarized in Table V.

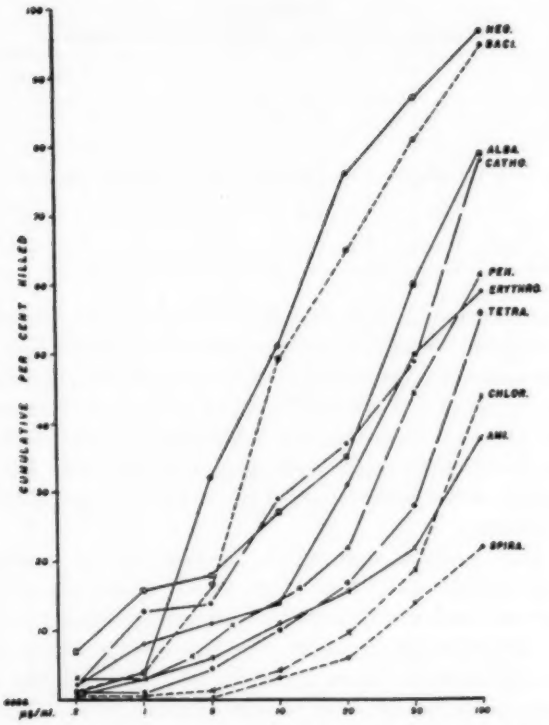

FIG. 1. The bactericidal effect of ten antibiotics on 68 strains of staphylococci.

Other Gram positive organisms.—Strains 69 to 75 (Table II) were isolated from severely ill patients, all of whom had bacteremia, with the exception of strain 71 which came from a brain abscess in a patient with cyanotic congenital heart disease. Two strains were highly sensitive to all agents (Nos. 69 and 72) and a third (No. 73) to all but penicillin. The growth characteristics of strain 70 were somewhat different; no agent except penicillin was *bactericidal* for this organism and all but novobiocin were *bacteriostatic*. The sensitivity pattern of this bacterium is similar to those previously reported for *Listeria monocylogenes* (3, 4). *Streptococcus fecalis* (No. 74) from the blood of a patient with subacute bacterial endocarditis was considerably more resistant than the other streptococci. Erythromycin was more consistently effective in this group than against the staphylococci tested. On the other hand, novobiocin was somewhat less effective in this group as a whole. Neomycin, tetracycline, and chloramphenicol *inhibited* bacterial growth in relatively low concentrations but could not be adjudged the drugs of choice against any of these organisms.

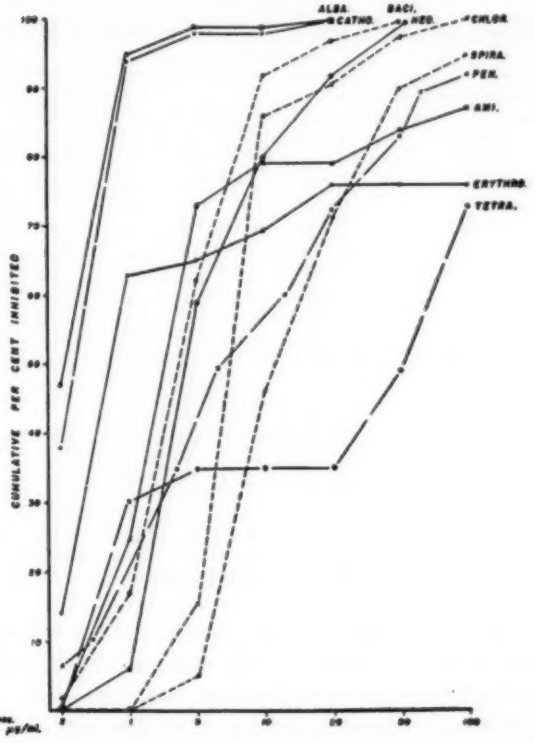

FIG. 2. The bacteriostatic effect of ten antibiotics on 68 strains of staphylococci.

Strains 75 to 80 were enterococci isolated from the urinary tract. They were uniformly resistant to novobiocin; spiramycin exerted only a mild *bacteriostatic* action.

Gram negative bacilli.—The detailed results of these tests are recorded in Table III. It may be seen that in almost every instance organisms proved to be resistant to one or more of the antibiotics. High concentrations of penicillin were bactericidal for a strain of *Paracolon* bacillus (No. 87) and a *Salmonella cholerasuis* (No. 89). Tetracycline was usually effective, *inhibitory* concentrations falling within the attainable therapeutic range (less than 20 mcgm./ml.) in nine of 13 tests. Five strains were totally resistant to chloramphenicol in all concentrations. Only neomycin and polymyxin were consistently *bactericidal* as well as bacteriostatic, but high concentrations were required before either effect was demonstrable. Streptomycin-resistance was common throughout the group. Novobiocin was uniformly ineffective in the 19 strains examined although a strain of *Proteus* (No. 97) manifested slight sensitivity to this agent.

TABLE V

Bactericidal and Bacteriostatic Activity of Nine Chemotherapeutic Agents Tested Against Staphylococci

Concentration of Drug u. or μg/cc.	.2	1	5	10	20	50	100	None
Erythromycin	1*	4	2	2	11	12	6	26
	9	31	2	2	5	0	0	15
Spiramycin	0	0	0	2	2	5	5	49
	0	0	3	26	16	12	3	3
Amimycin	0	2	2	3	3	4	10	39
	0	16	30	4	0	3	2	8
Cathomycin	1	7	1	9	5	8	18	14
	24	35	3	0	1	0	0	0
Albamycin	5	6	1	6	6	17	12	15
	32	32	3	0	1	0	0	0
Bacitracin	0	2	9	20	10	10	9	3
	1	10	28	19	3	2	0	0
Neomycin	0	2	18	12	16	7	6	2
	0	10	27	13	8	5	0	0
Tetracycline	0	0	3	4	4	7	17	28
	0	19	3	0	0	9	15	17
Chloromycetin	0	0	1	2	3	6	16	35
	0	0	10	44	4	4	1	0

* The first line is the number of strains killed, and the second, the number of strains inhibited.

The apparent lack of effectiveness of novobiocin on *Proteus* is somewhat contrary to other reports (5–7); however, this was a relatively small number of selected organisms from patients with severe infections.

Comparison of tube dilution sensitivities with the disk method.—The minimal *inhibitory* concentrations of antibiotics for staphylococci were compared with results obtained by the disk method (Table VI). The 68 strains studied in this series were compared with 211 successive strains isolated and tested in the routine clinical bacteriology laboratory of the Johns Hopkins Hospital during March, 1956. The criteria employed in the comparison are shown in Table VI (8) and the results in Table VII. Several points should be noted:

(1) The incidence of penicillin resistance was much higher in the selected series reviewed here than in the successive routine cultures for the entire hospital during a single month, taking 1.0 unit/ml. of penicillin as the dividing line.

(2) Resistance to tetracycline was approximately the same in both groups.

(3) Chloramphenicol's effectiveness was approximately the same in the two groups.

(4) The incidence of strains resistant to erythromycin was approximately three times as high in the selected as in the unselected group.

(5) Using the standards established for erythromycin's effectiveness *in vivo*,

TABLE VI

*Correlation Between Minimal Inhibitory Concentration of Drug and Sensitivity**

Drug	Sensitive	Mod. Sensitive	Resistant
Penicillin	≦.5 u.	1 u.	≧5 u.
Tetracycline	≦5 mcg.	10 mcg.	≧20 mcg.
Chloromycetin	≦10 mcg.	20 mcg.	≧50 mcg.
Bacitracin	≦1 u.	5 u.	≧10 u.
Neomycin	≦5 mcg.	10 mcg.	≧20 mcg.
Erythromycin	≦1 mcg.	—	≧5 mcg.
Spiramycin	Same as Erythromycin.		
Amimycin	Same as Erythromycin.		
Albamycin	Same as Erythromycin.		
Cathomycin	Same as Erythromycin.		

* From: Grove, D. C. and Randall, W. A. *Assay Methods of Antibiotics*, Pp. 192.

TABLE VII

Sensitivity of Staphylococci to Antibiotics Using Criteria in Table VI

Agent	No. Strains Sensitive Per Cent		No. Strains Mod. Sensitive Per Cent		No. Strains Resistant Per Cent	
	Present Series	JHH Bact. Lab	Present Series	JHH Bact. Lab	Present Series	JHH Bact. Lab
Penicillin	6.3	48.3	3.2	—	90.5	51.7
Tetracycline	34.9	46.4	0	—	65.1	53.6
Chloromycetin	87.3	92.9	7.9	—	4.8	7.1
Bacitracin	17.4	ND*	44.4	—	38.2	ND
Neomycin	60.3	ND	19.1	—	20.6	ND
Erythromycin	65.6	87.7	—	—	34.4	12.3
Spiramycin	4.8	ND	—	—	95.2	ND
Amimycin	71.4	ND	—	—	28.6	ND
Cathomycin	98.4	ND	—	—	1.6	ND
Albamycin	98.5	ND	—	—	1.5	ND

* Not done.

novobiocin was by far the best inhibitory agent and spiramycin was essentially impotent. Amimycin's position was an intermediate one on this scale.

DISCUSSION

The changing patterns of sensitivity of pathogenic bacteria to antibiotics during recent years has been repeatedly described and is well-documented; it requires no reiteration here. Emphasis has been placed upon the importance of staphylococci in many studies (9–11), but similar increases in resistance have been recognized for many other species. The data presented here represent a segment of the experience in a single hospital and certainly confirm, indeed exaggerate, the observations on staphylococci. Many of the strains were highly resistant to both penicillin and erythromycin. The pattern of the results with

penicillin is such as to suggest that this antibiotic is almost useless in staphylococcal infections. Actually, however, 77.8 per cent of the organisms were *inhibited* by 50 units/ml. of penicillin and 30 per cent were killed by that concentration. Intravenous infusion of large doses of penicillin coupled with the use of Benemid renders such concentrations attainable in clinical practice and it can be said that in many cases massive amounts of penicillin now produce results similar to those accomplished with small amounts a few years ago.

Despite some opinions to the contrary (12, 13), the present study certainly suggests that with the passage of time, more and more erythromycin resistant strains will appear. The fact that organisms were either inhibited by relatively small amounts of this drug or were completely unaffected contrasts with the rather uniform spread of penicillin's action in the present series.

The potent bactericidal action of neomycin and bacitracin makes these drugs extremely useful in the local treatment of infection and, indeed, despite their toxicity, they are coming to play an increasingly important role in systemic therapy.

The rather consistent effectiveness of chloramphenicol as an *inhibitory* agent for the staphylococcus observed in this group of organisms is in accord with many other reports (11, 13). However, only a rare strain is *killed* by this drug in concentrations that are usually obtained clinically.

This study offers little evidence that spiramycin will be of material value in the treatment of staphylococcal or streptococcal infections. Although it has been stated that this agent is more effective *in vivo* than *in vitro*, further clinical confirmation is needed (14).

The striking consistency with which novobiocin, in relatively low concentrations inhibited the growth of staphylococci is encouraging. With many strains, far higher amounts were needed for direct *killing* but clinically attainable concentrations of novobiocin were bactericidal for about 20 organisms. Certainly this drug promises to be a potent addition to the therapeutic armamentarium against staphylococci.

Oleandomycin was less active than novobiocin and a number of strains were completely resistant to its action *in vitro*. While it may eventually prove to have some limited usefulness, it was not as impressive in the present tests as several other agents.

The number of Gram negative bacilli studied is so small that few conclusions are possible other than the general one that this group is now responsible for many of the most difficult problems in clinical management of infection.

Lastly, it should be pointed out that only the results of clinical trials can provide the final answer to the usefulness of an antibiotic. Sensitivity tests, no matter how carefully carried out, can serve only as a relative guide and careful correlation of the results of both laboratory and clinical observations is of ut-

most importance. Such an analysis of the clinical outcome of the infections from which the present series of organisms was isolated is now in preparation.

SUMMARY

One hundred pathogenic organisms, including 68 staphylococci and 20 Gram negative bacilli, isolated from patients with severe infections, were tested against several antibiotics, including oleandomycin, spiramycin, and novobiocin. Determinations of both bacteriostatic and bactericidal concentrations were carried out and the results are discussed from the point of view of their implications for the clinical usefulness of the drugs tested. Particularly to be emphasized are the continuing effectiveness of penicillin in high dosage and the consistent bacteriostatic potency of novobiocin for staphylococci.

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FATAL HYPERTENSIVE CRISIS FOLLOWING DENERVATION OF THE CAROTID SINUS FOR THE RELIEF OF REPEATED ATTACKS OF SYNCOPE

Case History

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The purpose of this note is to call attention to the fact that in cases of carotid sinus syncope, denervation of the sinus may cause a fatal hypertensive crisis. I have the impression that this particular reaction is not generally recognized as one of the hazards of the operation.

Case History (No. 605484—Path. 23683) Mr. A.O., a man of 49 years, was seen in consultation with Dr. Lay Martin on May 12, 1952. He stated that in 1932 he had had some episodes with loss of consciousness of brief duration. These were preceded by a severe, brief pain in the left ear. After about 3 years these attacks ceased, but in 1940 they returned and persisted thereafter. In these episodes there was no pain in the ear and the loss of consciousness occurred without warning. It was found that stimulation of the left carotid sinus would induce the attacks at once. Atropine and other drugs were tried without result. The patient was advised not to wear a collar and not to turn his head suddenly. There was no relief. The attacks occurred without apparent cause. They became more frequent and more severe. Convulsive movements were seen in some. When the patient was seen in May 1952, he stated that in the preceding 12 months he had had 40 attacks of syncope. Nothing else of importance was elicited.

On examination the patient was well developed and nourished. The physical examination revealed nothing of importance. The neurological examination was completely negative. Blood pressure was rather labile ranging between 120/80 and 165/90. The pulse varied between 78 and 90. The temperature was normal. The blood counts were all normal and the blood sugar, non-protein nitrogen and serum electrolytes were within normal limits. The serological test for syphilis was negative.

The left carotid sinus was stimulated several times. The heart beat was always slowed or stopped. The patient lost consciousness for periods of 10 to 30 seconds. In some instances there was twitching of the right hand and of both legs. When the patient regained consciousness his mind was clear at once. It was found that atropine and other drugs were not helpful. It was decided that operation was indicated.

On May 27, the patient was given pentothal anesthesia with ether in addition. The blood pressure was 120/80. The pressure fell as the pentothal was given. The left carotid sinus was then infiltrated with procaine. At this point the blood pressure began to rise. The operation was carried out nevertheless and the left carotid artery was stripped of its sheath. At the end of the operation which required 45 minutes, the blood pressure had reached 240/140. It should be said that no mass or other anatomical abnormality was found at operation.

After operation the patient failed to regain consciousness and remained in coma. The blood pressure fell to 150/90 within a few hours after the operation. The lips were cyanotic. The musculature was flaccid and the tendon reflexes absent. The spinal fluid contained 40 red cells but no white cells. There was a trace of globulin. The pressure was 220 mm. water. A transfusion was given.

The patient did not improve. On May 28, the pulse grew weak and respiration shallow and irregular. Respiration ceased.

At post-mortem examination, the walls of the left ventricle were thickened. The coronary arteries showed arteriosclerotic plaques but were not occluded. The aorta showed arteriosclerotic changes. The lungs, kidneys, liver and other organs showed no lesions. The left carotid artery was grossly normal.

There was some flattening of the convolutions of the cerebral cortex, a small pressure cone about the cerebellum and signs of slight tentorial herniation. On section hemorrhages were found in the cortex of the left frontal lobe, left caudate nucleus, right internal capsule, and thalamus. The cerebral arteries showed arteriosclerotic plaques but were not occluded.

It seems probable that the hemorrhages occurred when the blood pressure was at its height and were due not only to the hypertension but to changes in the cerebral vessels which could not sustain the high pressure.

Discussion

It is well known that *excitation of the carotid sinus reflex* at operation may cause fatal cardiac arrest. Because of this danger the carotid sinus is routinely infiltrated with procaine before operation.

The danger of sudden *abolition of the carotid sinus reflex*, however, does not seem to be generally appreciated. Yet Bucy pointed out in 1936 that section of one glossopharyngeal nerve, through which the carotid sinus nerve reaches the brain, would cause elevation of blood pressure. He reported 5 cases in which one glossopharyngeal nerve was sectioned because of ninth nerve neuralgia. The section was made intracranially. In 4 out of 5 cases there was prompt and pronounced rise of blood pressure. This hypertension lasted for from 5 to 12 days. In one instance the pressure rose from 120/90 to 186/80 in 7 minutes and in another case from 114/72 to 190/120. Bucy points out that the carotid sinus mechanism gives rise to a constant flow of nervous impulses which have

the effect of reducing the blood pressure and sudden interruption of these impulses causes a transient hypertension before other mechanisms can gain control.

In our patient the blood pressure rose from 120/80 to 240/140 in a short time causing a fatal hypertensive crisis with multiple cerebral hemorrhages.

In describing the effects of roentgenotherapy in cases of carotid sinus syncope Greeley *et al.* do not mention hypertension. Possibly the reflex is reduced gradually in such instances and hypertension does not occur. The syncope was stopped in only 58 per cent of cases by this method of treatment, however.

CONCLUSION

1. Denervation of the carotid sinus on one side in cases of carotid sinus syncope may cause a severe and fatal hypertensive crisis. 2. Hypertension due to section of the ninth nerve reaches its peak in a short time and then gradually declines to its previous level within 5 to 12 days. 3. It is possible that this hypertensive reaction may be avoided when roentgen irradiation is employed because the reflex is reduced gradually. No doubt, the hypertensive reaction may be controlled at operation by the use of hypotensive drugs.

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THE RATE OF PENICILLIN'S BACTERICIDAL ACTION ON STAPHYLOCOCCI *IN VITRO*; SOME OBSERVATIONS ON THE SO-CALLED ZONE PHENOMENON[1]

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It has been known for a number of years that the time required for penicillin to kill bacteria in an *in vitro* system is not necessarily shortened by increasing the concentration of antibiotic beyond a certain "optimal" level. The experiments of Eagle and Musselman (1) showed clearly that with most species, the addition of penicillin to cultures in concentrations exceeding this optimum simply results in a plateau of maximal drug action. Of perhaps greater biological interest, however, and certainly a fact that could have great clinical importance, is the finding that under certain conditions the presence of an excess of penicillin can result in a less efficient bactericidal action. Such a situation was described by both Kirby (2) and Garrod (3, 4) and during Eagle's extensive studies of the factors influencing penicillin's action, he found that some strains of streptococci and staphylococci survived for a longer time in high than in low concentrations of penicillin (5–7). This paradoxical reduction in the bactericidal rate of penicillin was termed the "zone phenomenon" by Eagle and has been suggested by a few observers as one explanation for failure of treatment in certain infections, especially bacterial endocarditis (8, 9). To select an organism that could be used to study the possible occurrence of a zone phenomenon in the treatment of experimental infections with penicillin, several strains of *Staphylococcus aureus* were tested for "zoning" *in vitro*. This report describes some findings in relation to the conditions under which a paradoxical action of penicillin is demonstrable *in vitro* and the incidence of staphylococcal strains that show this type of response.

MATERIALS AND METHODS

Sources of cultures.—Eight strains of *S. aureus* (designated A, H, I, K, L, M, O, and R), isolated from patients with serious infections ranging from endocarditis to localized abscesses, were selected. All had been shown, by tube-dilution sensitivity tests, to be inhibited by less than 1.0 unit of penicillin/ml. In addition, the "Smith" strain of *S. aureus*, kindly supplied by Dr. Harry Eagle, was tested. All organisms were stored on agar slants at 4°C. until use, at which time a single colony from the slant was inoculated into 10 ml. of infusion broth (Baltimore Biological Laboratories), incubated for 18 hr. at 37°C., and stored at 4°C. as the *stock culture* for all experiments.

[1] This study was supported by a Contract with the Army Chemical Corps, Fort Detrick, Maryland.

Preparation of Inoculum.—As will be detailed under "Results", the method of preparing the *inoculum* was found to be of crucial importance in testing for the zone phenomenon with penicillin. The following procedure was used: a loopful of the stock broth culture was mixed with 2.5 ml. of infusion broth and incubated at 21°, 30°, or 37° for a specified period of time. Best results were obtained with inocula incubated at 30°C. for exactly 14 hr. and exposed immediately to the antibiotic.

Method of testing.—Solutions of crystalline penicillin G in concentrations of 0.2, 0.5, 1.0, 1.5, and 200 units/ml. in infusion broth were prepared in quantity and stored at −20°C. A volume of 0.1 or 0.2 ml. of the *inoculum* to be tested was added to each of a series of tubes containing 4.8 or 4.9 ml. of the various concentrations of penicillin in broth so that the final volume was 5.0 ml. After thorough mixing, cultures were incubated at 37°C. Aliquots for bacterial counts were taken from each tube immediately and at 3, 6, 12, and 24 hours. Samples were diluted serially with sterile saline and replicate pour-plates were made using nutrient agar to which had been added penicillinase (Schenley). After incubation at 37°C. for 32 hr., colony counts were performed, using a Quebec counter. Several preliminary attempts to utilize turbidimetry as described by Kirby (2) yielded variable results, perhaps because of the smaller inocula and shorter periods of observation employed in the present study.

RESULTS

Influence of time and temperature of incubation of the inoculum upon the zone reaction.—In preliminary experiments, the eight strains of *S. aureus*, A through R, were tested by the method described above, using inocula that had been incubated overnight at 30°C. or 37°C. This procedure was used primarily because of its convenience and because it approximated the schedule described by Eagle and Musselman (1) who employed cultures incubated at 30°C. for 15 hours. With none of the organisms tested was there any indication of impairment or slowing of bactericidal activity in the higher concentrations of penicillin. Indeed, killing was consistently more rapid in the tubes containing the largest amounts of the antibiotic.

Failure to find any zone phenomenon in tests with inocula prepared by incubation at 37°C. was not wholly unexpected. Verwey (10) has reported that several strains of bacteria that showed a paradoxical reduction in susceptibility to the bactericidal activity of high concentrations of penicillin when tested after incubation at 30°C. demonstrated no evidence of any zone reaction when retested after incubation at 37°C.

However, the absence of a zone reaction with inocula incubated at 30°C. in the present study was somewhat more difficult to explain and seemed to indicate that none of the eight strains of staphylococcus selected for testing possessed the particular property of longer survival in high concentrations of penicillin than in low concentration. Although Kirby (2), whose technique differed in several respects from that of later investigators, had noted a zone reaction with all strains of staphylococci studied, the reports of Garrod (3, 4) give the impression that this type of reaction was observed by him with some, but not all, organisms tested. Furthermore, of nine staphylococci tested by

Eagle and Musselman (1) two showed a pronounced zone effect with penicillin, two others a slight, but definite zone, and five, none at all. To check on this explanation and also to evaluate the technique employed in the present study, the "Smith" strain of *S. aureus* was obtained from Dr. Eagle. This was one of the strains shown by Eagle and Musselman (1) to survive for longer periods in high concentrations of penicillin than in low. The Smith strain was tested several times, using inocula incubated overnight at 30°C., and in no instance was the zone phenomenon observed.

Re-examination of experimental protocols revealed that the period of "overnight" incubation that had been used in all of the tests thus far described had always exceeded 15 hours, usually amounting to 16–17 hours. All nine strains of staphylococci were therefore retested by exposing cultures that had been incubated for exactly 15 hours at 30°C. to the several concentrations of penicillin. Under these conditions, three of the strains (I, L, and Smith) showed distinct zone reactions, the rate of killing being consistently slower in tubes containing 200 units/ml. of penicillin than in those containing less than 2.0 units/ml.

When all staphylococci were again tested, employing inocula incubated at 30°C. for exactly 14 hours, striking impairment of the rate of killing in 200 units/ml. of penicillin was consistently demonstrable with every strain. Furthermore, strains A, H, I, and K were tested by exposure to penicillin after incubation at 21°C. for 20 hours and, again, all four showed clear-cut zone reactions.

From these and similar studies employing varying periods of incubation of inocula before exposure to penicillin, it became obvious that under the conditions of these experiments, the demonstration of a paradoxical increase in survival time of staphylococci in the presence of large amounts of penicillin was exquisitely dependent upon time and temperature of growth *before* exposure to the antibiotic. When a temperature of 30°C. and an incubation time of 14 hours were used, the zone phenomenon was easily elicited with all strains of staphylococcus tested. Once penicillin had been added to a culture, the incubation temperature appeared to have no influence on the results and at 37°C. or 30°C., the zone reaction was clearly evident.

The results of exposing staphylococci to varying concentrations of penicillin.—Exposure of an appropriately prepared culture of staphylococci to low and high concentrations of penicillin in broth followed by incubation at 37°C. resulted in more rapid diminution of bacterial counts in tubes containing small amounts of the antibiotic. This difference was sometimes evident within three hours, was invariably present within six hours, and became striking at 12 and 24 hours.

Table I shows the proportion of the original inoculum remaining when each organism was exposed to two low concentrations and a single high concentration

TABLE I

*Number of Viable Staphylococci Remaining in Broth Cultures at Various Intervals After Exposure to Penicillin at 37°C.**

Strain	No. Bacteria/ml. Orginal Inoculum (×10⁶)	Penicillin (units/ml.)	Percentage of inoculum viable after:			
			3 hr.	6 hr.	12 hr.	24 hr.
A	3.68	0.5	93	11.2	0.28	0.37
		1.0	14	3.5	0.078	0.0018
		200.0	11.6	6.9	1.8	0.019
H	3.44	0.2	282	326	16	38
		0.5	22.7	0.9	0.04	0.017
		200.0	55.4	10.2	2.7	0.208
I	5.06	1.0	58.0	8.0	0.10	0.286
		1.5	58.6	13.3	0.15	0.003
		200.0	46.2	21.5	1.58	0.069
K	5.12	0.2	170	36	0.9	97.0
		1.0	44	7.7	0.5	0.031
		200.0	67	26	3.8	0.138
L	7.15	0.2	63.5	6.5	0.008	0.185
		0.5	52.4	8.75	0.8	0.0024
		200.0	25.8	10.0	2.1	0.0034
M	3.40	0.2	45	0.26	0.0029	0.0
		0.5	3.7	0.036	0.0	0.0
		200.0	30	2.4	0.088	0.0
O	1.28	0.5	41	6.1	0.23	12.2
		1.0	26	9.9	0.49	0.049
		200.0	35	19.8	1.9	0.061
R	3.70	0.2	59	2.8	0.052	0.42
		0.5	8.4	0.43	0.072	0.005
		200.0	55	7.90	0.22	0.041

* All inocula prepared by incubation at 30°C. for 14 hours.

of penicillin. No attempt was made to establish the true "optimal" concentration of antibiotic for each strain and it can be seen that with the exception of strain M the smallest concentration allowed more organisms to survive after 24 hours than did the high concentration of 200 units/ml. However, the tubes containing slightly higher concentrations of penicillin uniformly contained fewer viable cells at 24 hours than 200 unit cultures. It is possible that intermediate concentrations of penicillin would have given an even more efficient bactericidal action, but this was not studied. It is interesting, however, to note that after 12 hours even the lowest levels of penicillin had killed more organisms than had 200 units/ml. and that reversal of this pattern during the 12–24 hour period was a result of multiplication of the remaining organisms, giving more "survivors" at 24 hours than at 12.

Figure 1 is a graphic representation of a typical penicillin zone phenomenon in a single test with strain A of *S. aureus*.

Table II summarizes the results of the tests depicted in Table I from the

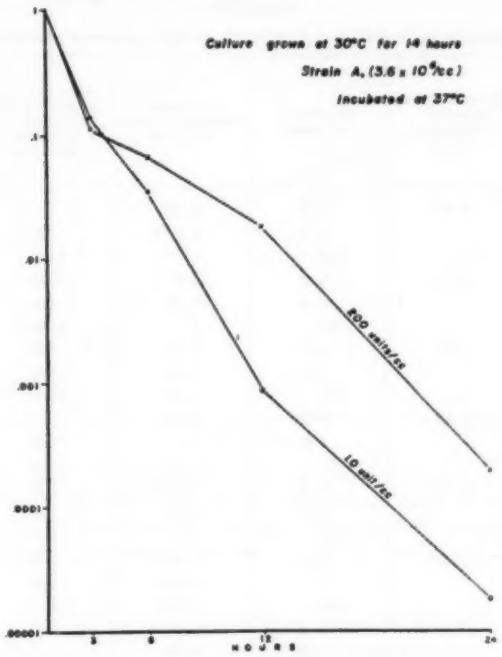

FIG. 1. Zone phenomenon with *Staphylococcus aureus* showing the more rapid killing by low concentration of penicillin.

point of view of time required for each concentration of penicillin to kill 99.9 per cent of the inoculum. With the exception of strains H and K, both low concentrations of antibiotic killed 99.9 per cent of the inoculum more rapidly than did 200 units/ml. although, because of renewed growth in the smallest amount of penicillin, an increase in viable organisms occurred during the interval between 12 and 24 hours.

The growth-stage of the inoculum and the zone phenomenon.—An attempt was made to investigate the repeated failures in preliminary experiments to obtain any evidence of the zone phenomenon with penicillin until cultures were prepared by incubation at 30°C. for 15 hours or, preferably, 14 hours before addition of the antibiotic.

The growth curves of four strains (A, H, I, K) were determined. A loopful of stock culture was inoculated into infusion broth and incubated at 30°C. At intervals of 1 to 3 hours, aliquots were withdrawn and nutrient agar pour-plates were prepared from serial dilutions in sterile saline. No penicillinase was added to the agar. With each of these organisms, it was found that multiplication in infusion broth proceeded in *two distinct exponential phases*, sepa-

TABLE II

Time Required for Penicillin to Kill 99.9% of Staphylococci Exposed at 37°C.

Strain	No. Bacteria/ml., Original Inoculum ($\times 10^9$)	Penicillin (units/ml.)	% Bacteria remaining viable after 12 hr.	Time required for penicillin to kill 99.9% of inoculum (minutes)	% of organisms viable when 99.9% of bacteria had been killed by the "optimum" level of penicillin
A	3.68	0.5	0.28	900	0.35
		1.0	0.078	700	0.10
		200	1.79	1140	2.0
H	3.44	0.2	16.0	2800	38.0*
		0.5	0.04	620	0.1
		200	2.7	1640	4.0
I	5.06	1.0	0.10	720	0.11*
		1.5	0.15	780	0.10
		200	1.58	1360	1.2
K	5.12	0.2	0.9		3.5*
		1.0	0.5	1120	0.1
		200	3.8	1500	0.6
L	7.15	0.2	0.008	600	0.04*
		0.5	0.8	940	0.10
		200	2.1	1060	0.23
M	3.40	0.2	0.003	440	2.4
		0.5	0.000	280	0.1
		200	0.088	580	5.4
O	1.28	0.5	0.23		3.8*
		1.0	0.49	1110	0.10
		200	1.90	1340	0.18
R	3.70	0.2	0.42	660	0.12*
		0.5	0.005	640	0.10
		200	0.041	860	0.48

* Logarithmic growth occurred in these tubes after the twelfth hour of exposure to penicillin.

rated by a brief *static phase* during which there was no detectable evidence of any increase in the number of bacteria. Such a growth curve for strain A is illustrated in Figure 2 in which the short lag period between two logarithmic growth periods is clearly shown. Of greatest interest was the fact that this transient cessation of multiplication occurred consistently between the 13th and 15th hours after inoculation, coinciding exactly with the time when exposure of the culture to penicillin resulted in the zone phenomenon.

The possibility that there existed some relationship between the static phase at the 14th hour and the zone phenomenon was tested by removing aliquots of the culture at various times during the period of incubation, exposing these samples to high and low concentrations of penicillin the usual way and testing for the zone reaction. The results were consistent and striking. Only inocula taken during the short static phase at the 14th hour were killed at a lesser rate by high concentrations of penicillin. Samples obtained during the first period

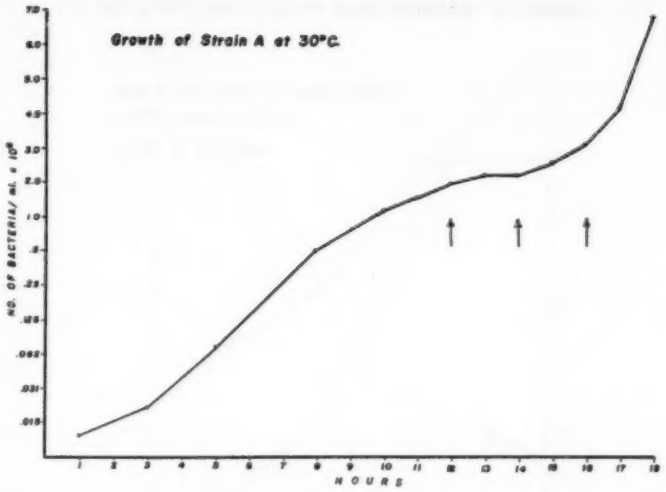

FIG. 2. Growth curve of *Staphylococcus aureus*, Strain A, in infusion broth at 30°C., showing the "di-auxie" effect consisting of two exponential phases of growth separated by a static phase at 13–15 hours. Note the change in scale along the vertical axis. The arrows indicate times samples were taken for exposure to penicillin (see Figs. 3–5).

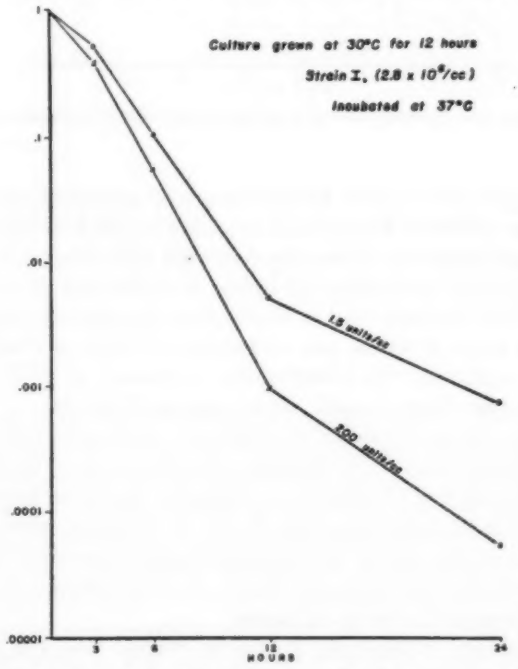

FIG. 3. Failure of culture of *Staphylococcus aureus*, Strain I, grown at 30°C. for 12 hours to show zone phenomenon with penicillin.

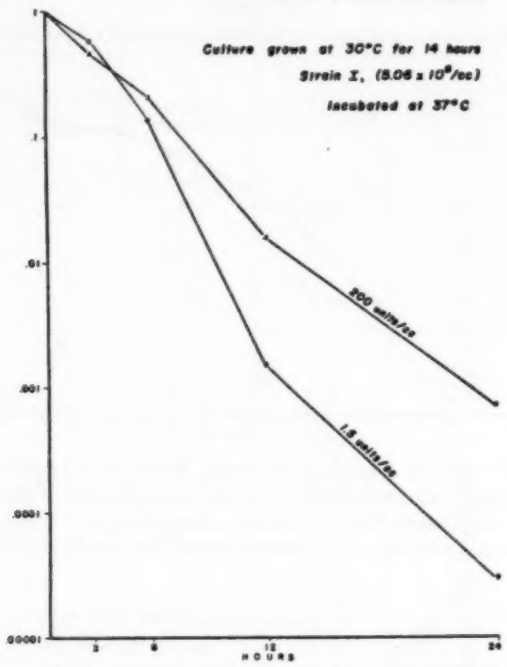

FIG. 4. Typical zone phenomenon with penicillin in culture of Strain I grown at 30°C. for 14 hours.

of exponential growth or later, during the second period of rapid multiplication, showed no evidence whatever of any paradoxical survival in the higher concentrations of penicillin. This type of finding with strain I is illustrated in Figures 3–5. Samples were taken 12 hours, 14 hours, and 16 hours after the start of incubation (as indicated in Figure 2 by the arrows). Each was inoculated into two tubes of broth, one containing 1.5 units of penicillin/ml. and the other 200 units/ml.; the cultures were incubated at 37°C. and periodic colony counts were made in agar containing penicillinase.

The inoculum taken at 12 hours was killed at a more rapid rate by 200 units/ml. of the antibiotic (Fig. 3). In contrast, with the inoculum taken at 14 hours, the bactericidal action of 1.5 units of penicillin/ml. was by far the more efficient. This typical zone reaction is shown in Figure 4. However, when the inoculum taken at 16 hours was tested, the organisms again responded to penicillin in the same manner that they had at 12 hours, the high concentrations producing a more rapid decrease in viable organisms.

From experiments of this type, it became evident that the demonstration of a paradoxical reduction in the rate of bactericidal action against staphylococci of high concentrations of penicillin in nutrient broth was dependent upon

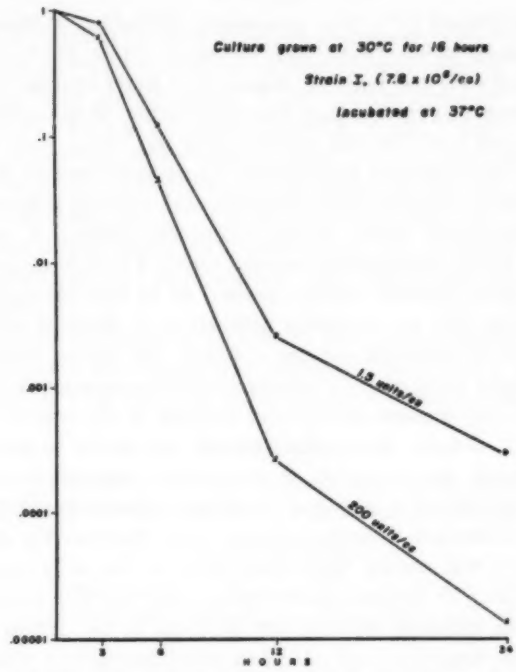

FIG. 5. Failure of culture of Strain I grown at 30°C. for 16 hours to show zone phenomenon with penicillin.

exposure of organisms to the antibiotic at a time when multiplication had undergone spontaneous temporary cessation. The number of organisms exposed and, as mentioned, the temperature of incubation after addition of penicillin were found to exert no appreciable influence upon the rate of killing when such an inoculum was used.

DISCUSSION

The present study was undertaken as a preliminary to investigations of the possible occurrence of a penicillin zone phenomenon *in vivo*. The obvious importance of information about a deleterious action of overdosage of penicillin in bacterial infections led to plans to select an organism that reacted paradoxically to penicillin *in vitro* and to produce infection in experimental animals. There were several reasons for choosing the staphylococcus. The recent increase in human infections caused by strains of this organism resistant to relatively large amounts of penicillin has led to clinical use of this drug in enormous dosages. Furthermore, the staphylococcus is often infectious for mice and satisfactory techniques for determining the bacterial content of various tissues

have been developed (11). The unforeseen difficulties in demonstrating the zone phenomenon *in vitro*, even with the Smith strain, led to an examination of factors influencing the reaction. When more exact conditions for preparing test inocula were observed, a paradoxical reduction in the rate at which high concentrations of penicillin kill staphylococci was found with regularity.

The finding that transient susceptibility to the zone reaction of staphylococci growing in infusion broth at 30°C. coincides with a period of spontaneous cessation of multiplication raises several interesting points. A growth pattern characterized by two exponential phases separated by a resting phase has been described in other cultures and is referred to as the "di-auxie" effect (12). Di-auxie is often due to successive utilization of different substrates necessitating the use of different enzyme systems. The pause between periods of growth is thought to represent a time of enzymic reorganization and metabolic readjustment. The present results offer nothing in the way of a biochemical explanation for di-auxie when staphylococci are grown in nutrient broth at 30°C; pH change, accumulation of catabolites, exhaustion of a nutritional source, or a combination of these "limiting factors" could be responsible. There is every reason to believe, however, that whatever the metabolic readjustment during the resting phase may prove to be, it is closely connected with susceptibility to the zone phenomenon and that this susceptibility is an exceptional and transient state in the life-cycle of the culture.

Eagle (7) has suggested that the zone phenomenon is a reflection of penicillin's ability to affect bacterial metabolism in more than one fashion. He postulates that in low doses, the antibiotic's lethal action could result from enzymic blockade at a stage that causes accumulation of a toxic metabolite. Larger doses may act at an earlier step in the reaction, preventing synthesis of the toxic substance. In other words, low doses produce death by "poisoning" and high doses by "starvation". Thus, while either metabolic lesion is eventually lethal, "poisoning" by low doses produces death at a more rapid rate. The present findings do not conflict with this hypothesis; indeed they may strengthen it by suggesting in addition that the metabolic milieu for this double lethal action of penicillin exists only transiently during growth *in vitro*. Certainly, studies aimed at biochemical characterization of penicillin's mode of action may profitably concentrate attention upon events during the static phase.

The results described here are preliminary. Confirmation and extension will be necessary before their applicability to other bacterial species or to events *in vivo* can be assumed.

Studies are now in progress to examine growth curves of staphylococci in other media, to examine zoning in other bacterial species in relation to the growth curve, and to determine the influence upon penicillin's action of artificial induction of a cessation of multiplication in a culture.

SUMMARY

A study of the decrease in rate of killing of staphylococci that occurs when these organisms are exposed to an excess of penicillin is reported. The occurrence of a "zone phenomenon" of the type described by Eagle was confirmed for each of nine strains tested. However, it was found that the demonstration of this paradoxical reduction in penicillin's bactericidal action is dependent upon exposure of organisms to the antibiotic during a brief period of spontaneous cessation of growth between phases of logarithmic multiplication. This transient susceptibility to the zone reaction occurs after 14 hours of incubation at 30°C. and lasts for about one hour when staphylococci are grown in infusion broth.

These results are discussed in relation to the zone phenomenon with other bacterial species, the biochemical mechanisms of penicillin's action, and the possible occurrence of a comparable prolongation of killing time *in vivo*.

ACKNOWLEDGMENT

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FIVE CASES OF AN UNDESCRIBED FORM OF PULMONARY INTERSTITIAL FIBROSIS CAUSED BY OBSTRUCTION OF THE PULMONARY VEINS

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In a brief period of two years, from February of 1952 to February of 1954, we were confronted with five cases which presented an unusual form of pulmonary interstitial fibrosis. It appeared highly significant that they all had as a common factor complete, partial, or perhaps intermittent obstruction of one or more pulmonary veins. In attempting to learn more about the association of interstitial fibrosis and pulmonary vein obstruction it was anticipated that a search of the literature would yield similar cases. To our surprise this search proved to be in vain; with one possible exception (1) to be referred to later, we were unable to find a single case in which pulmonary interstitial fibrosis was described in association with pulmonary vein obstruction. Indeed, there was even a surprising paucity of reported cases with demonstrated obstruction of the pulmonary veins from any cause, and in these few cases a description of the lung drained by the obstructed vein was either entirely inadequate or completely lacking. The hitherto unrecognized association of pulmonary vein obstruction and interstitial fibrosis, together with the evidence provided by our cases that the interstitial fibrosis was the result of pulmonary vein obstruction, are the reasons for the present report. The cases will be described in the order in which they came to autopsy.

Case # 1

(Hist. No. 593271). This 33 year old white housewife was admitted to the Johns Hopkins Hospital for the first and only time on December 16, 1951, for "rheumatic heart disease and intermittent fever".

Since early childhood, she had had frequent "sore throats", often accompanied by "growing pains". At age 16 she was first told that she had a heart murmur. When she was 25 she had another "sore throat" and with this she had for the first time swollen, red, painful joints. She had two pregnancies, one at age 27 and another when she was 29; both were accompanied by hypertension, dyspnea and ankle edema. Sometime after the second pregnancy easy fatigability, cough, orthopnea, and exertional dyspnea appeared and became persistent. A year before admission the patient had a transient episode of jaundice. Several months before admission she developed intermittent fever. This was often associated with cough, chest pain and increasing dyspnea, and on several occasions required hospitalization. Antibiotic and other therapy became increasingly less effective. Chest x-rays during this time revealed "haziness" in the right lower lung field. A 60 lb. weight loss had been noted.

On admission to this hospital her temperature was 101, pulse 100, respirations 20 and blood pressure 100/80 mm. Hg. There was no cyanosis, edema, or "clubbing". The lungs were clear to percussion and auscultation except, posteriorly and laterally, over the right base where percussion was impaired and the breath and voice sounds, as well as tactile fremitus were decreased. Fine crackling rales were also heard in this area. The heart was slightly enlarged to the left. Its rate was rapid but regular. P2

was greater than A2. At the apex there was a loud, high pitched, cooing murmur, which occurred early in systole and was poorly transmitted. At the base of the heart and just to the right of the sternum there was a low pitched, softer systolic murmur, which was also transmitted poorly. Diastolic murmurs were not heard. Except for a markedly enlarged liver, examination of the abdomen was negative. A white count of 13,440 and a corrected erythrocyte sedimentation rate of 20 mm./hr. were the only abnormalities on routine blood and urine examinations. Innumerable blood and sputum cultures were taken at various intervals and all were negative. Radiological examinations of the chest revealed enlargement of the right auricle and ventricle, as well as marked left auricular enlargement. Throughout her hospital course, an unchanging mottled infiltrate or haziness was present in the right lower lobe. Electrocardiograms consistently showed right axis deviation and right ventricular "strain". Chief among the many problems presented by her illness was the persistent, although intermittent fever. This was never satisfactorily explained, and in general it responded rather poorly to vigorous antibiotic therapy. With minor fluctuations all of the signs and symptoms present on admission persisted throughout her hospital course. Various diagnoses were suggested; the most popular seemed to be mitral stenosis with bacterial endocarditis. Various forms of therapy were, for the most part, unsuccessful. She gradually deteriorated, until terminally her respirations became very rapid; her temperature rose to 106°; she went into circulatory collapse and developed intractable pulmonary edema. Death occurred on the 51st hospital day.

At *autopsy* (# 23466) the heart was enlarged, weighing 410 grams. There was considerable dilatation and hypertrophy of both the right auricle and right ventricle. The left auricle was also markedly enlarged, and when it was opened a huge gelatinous tumor, measuring 8 x 7 x 6 cm. was found, virtually filling the entire chamber. One end of the tumor was attached to the interauricular septum, while the other end floated free and extended to the orifice of the mitral valve. It was not until considerably later that the importance of pulmonary vein obstruction in this case was fully appreciated, and it was then too late to study pointedly the relation of the tumor to the orifices of the individual pulmonary veins, although it was clear that the flow from any of these orifices might have been embarrassed by this huge tumor. Grossly and microscopically, the tumor was a typical myxoma. All of the valves, including the mitral, were normal. There was nothing to suggest rheumatic fever in any of the microscopic sections of endocardium, valves, or myocardium, nor was there any evidence of bacterial endocarditis. As an explanation for the clinical picture of bacterial endocarditis in this case, it should be mentioned that we have seen other cases in this hospital which clinically presented a fairly typical picture of bacterial endocarditis, and at autopsy revealed an auricular myxoma and no evidence of bacterial endocarditis. A number of similar cases have been reported in the literature (16). The *lungs* were unusually heavy, weighing approximately 2000 grams. On section, they were diffusely edematous. In addition, the right middle and lower lobes, as well as scattered patches in other lobes, were gray and indurated. This induration appeared to be the result of an interlacing network of fibrous tissue which was outlining readily discernible alveoli. The liver was markedly congested, both grossly and microscopically. There were no significant changes in other organs.

Microscopic sections from the gray, indurated areas of lung disclosed, in essence, a bland interstitial fibrosis (Fig. 1). The alveolar walls in these areas were greatly thickened by a relatively acellular lacework of mature collagen fibers. The resultant alveolar changes were of some interest. There was great variation in their size; the majority were lined by a low cuboidal epithelium, and, except for occasional heart failure cells, they were invariably empty. Sections from the non-indurated areas of lung revealed in general little more than the ordinary picture of severe chronic passive congestion. However it was of considerable interest that in many of these areas there was slight, although definite fibroblastic proliferation within the alveolar walls. The small and medium-sized pulmonary arteries in the areas of interstitial fibrosis were often markedly narrowed, both by medial hypertrophy and intimal proliferation. In fact, one could not help but be reminded of an obliterative endarteritis, although it was, of course, appreciated that recanalized thrombi could give an entirely similar picture. Furthermore, it should be stressed that in the relatively unaffected or non-indurated areas the arteries were not appreciably narrowed, nor were fresh or old thrombi seen in the many examined.

Comment: The significance, in this case, of an unusual form of pulmonary interstitial fibrosis and its association with a left auricular myxoma large

enough to interfere with the flow of blood from the pulmonary veins was not fully appreciated until after study of the following cases.

Case #2

(Hist. No. 594258). A 37 year old white housewife was referred to the Johns Hopkins Hospital and admitted on March 21, 1952, for "rheumatic heart disease and possible commissurotomy". Although this woman had had typical acute rheumatic fever when she was twelve years old, there were apparently no signs or symptoms of heart disease until her first pregnancy, at age 25, when she was told that she had a "bad and leaky heart" and should have no more children. It was not until six or seven years later that exertional dyspnea and easy fatigability finally appeared. She was then admitted to a hospital and digitalized. Following discharge she continued taking digitalis, but remained a mild "cardiac cripple" throughout the next 3 or 4 years. During this time her dyspnea continued and she had two hemoptyses. There had never been any significant cough. Three months before admission she visited the Diagnostic Clinic of this hospital where it was felt that, with one exception, her signs and symptoms had been, and were, quite classically those of rheumatic mitral stenosis. That exception was the presence of an atypical, musical, systolic murmur. She returned home, but while awaiting admission for more complete evaluation, developed fever, malaise and a grippe-like illness. She was hospitalized elsewhere, treated with antibiotics and improved rapidly, but remained in that hospital until she was transferred here.

On admission, there was an irregular pulse and a low-grade fever. The neck veins were distended. There was no edema, cyanosis or "clubbing". The lungs were clear. A systolic thrill was palpable over the whole precordium. There was a diastolic shock in the pulmonic region. The heart was enlarged to the right and an inconstant, rough, loud, harsh systolic murmur was heard over the entire precordium and between the scapulae posteriorly. This seemed to be quite different from the "atypical murmur" heard 3 months previously in the Diagnostic Clinic. A diastolic murmur was also heard at the apex and along the left sternal border. P2 was louder than A2. The abdomen was full. Shifting dullness and a fluid wave were elicited. The liver seemed slightly enlarged. Except for a corrected sedimentation rate of 27, hematological studies were normal. Auricular fibrillation and right ventricular strain were demonstrated by electrocardiogram. The venous pressure was 228 mm. of 5 per cent glucose. Radiological examinations of the chest indicated an enlarged right ventricle, a prominent pulmonary conus and a very large left auricle. The bronchovascular markings were accentuated bilaterally. During the first week in the hospital, the history, the intermittent fever and the apparent change in murmurs all suggested mitral stenosis and bacterial endocarditis. Since she had been receiving penicillin before admission to this hospital eleven negative blood cultures did not seem surprising. During the second week her temperature went even higher and there were increasing signs of heart failure. On the 15th and 16th hospital days the patient had several peculiar episodes of shock with cardiac arrest. The possibility of an intra-auricular mass was suggested and it was felt that radical postural maneuvers might be of some help both diagnostically and therapeutically, therefore as a last resort the patient was held by the feet and literally shaken with her head down. With this maneuver the heart beat returned, but at the same time she developed pulmonary edema; this was partially cleared by returning the patient to the sitting position. However, shock and cardiac arrest soon returned and even though this maneuver was repeated, it and conventional measures finally failed and she died on the sixteenth day.

At *autopsy* (#23590) there was slight pitting edema of the lower legs and marked cyanosis of the head and neck. There were small pleural effusions, but no ascites. The heart was enlarged, weighing 460 grams. The right ventricle and left auricle were dilated and hypertrophied. On opening the left side of the heart a large pedunculated mural thrombus was found practically filling the auricular chamber. It measured 7 x 3 x 3 cm. It was attached only to the superior and lateral wall of the left auricle, while the tip, which floated free, extended into the orifice of the stenotic mitral valve. As in the preceding case, the relationship of pulmonary fibrosis to obstruction of the pulmonary veins was not appreciated at the time of autopsy, and therefore no pointed study of the positions of the thrombus in relationship to the orifices of the pulmonary veins was made, although again it was perfectly clear from the size of the thrombus that any, or all of them might have been, at least intermittently,

obstructed by this huge, mobile mass. Microscopic study of the thrombus was of interest only in that most of it was quite old; near its tip the thrombus was more recently formed and it was this fresh portion of the thrombus which had extended into the stenotic orifice of the mitral valve, and which may have accounted for the rapid progression of events terminally. In addition to the marked thickening and stenosis, the mitral valve also had short, scarred chordae tendineae. The left auricular endocardium was also thick and had a rough, granular surface. There was definite, although slight, thickening of the tricuspid valve and its chordae tendineae. The aortic and pulmonary valves were normal as was the endocardium of each ventricle. Sections of the heart showed perivascular scars with the occasional remains of an old Aschoff body. The myocardial fibers of the left auricle and right ventricle were hypertrophied, and the lining of the former showed thick layers of hyaline scar with foci of calcification. Even though edema was diffuse and definite, the lungs weighed only 980 grams. On section, they were firm but not consolidated. There was some rusty pigmentation. The liver presented the typical "nutmeg" appearance of severe chronic passive congestion, and, in addition, microscopic examination revealed early cardiac cirrhosis. There were no other relevant gross abnormalities.

Microscopically, the changes in the lungs of this case were not as extensive as those of the preceding case (Fig. 2). However, it was clear in fairly large, although widely scattered areas in both lungs that something out of the ordinary had happened to the alveolar walls. They were strikingly thickened by fibroblastic proliferation to an extent rarely seen, even in the most protracted and severe cases of mitral stenosis. In fact, there were a few areas in which the fibrotic process was becoming mature and relatively acellular, entirely like that of the preceding case. Similarly, medial hypertrophy and intimal thickening had severely narrowed many arteries and arterioles in these fibrotic areas. Rarely, a vessel would show a fresh arteritis, consisting mostly of infiltration of its wall by inflammatory cells. This process is rarely, if ever, in our experience associated with mitral stenosis unless active rheumatic fever (fresh Achoff bodies) is present, although it has been associated with severe pulmonary hypertension from other causes (2). Scattered foci of "heart failure" cells and edema coagulum were seen in all sections.

Comment: In this case, pulmonary interstitial fibrosis was found in association with a left auricular thrombus large enough to impinge upon the orifices of the pulmonary veins.

Case #3

(Hist. No. 600747). A 42 year old white housewife was admitted to the Johns Hopkins Hospital for the first time in March of 1952, complaining of "shortness of breath and spitting up blood". Her second and final admission was in October of the same year. The onset of the present illness apparently occurred in 1943, when at age 32, the patient coughed up blood-tinged sputum. During the next few years she continued to have occasional hemoptyses and also developed a dry, hacking cough. In 1947 she had a copious hemoptysis and also experienced for the first time slight exertional dyspnea and some palpitation. She was studied in Philadelphia at this time and again in 1948. She was told that she had mitral stenosis and was digitalized. The latter proved ineffective and was discontinued five weeks later. By 1950, dyspnea had progressed until it was present even on slight activity, and her hemoptyses had also increased both in frequency and in quantity. In December of 1950, and again in October of 1951, she was studied in Boston. On those occasions her heart was slightly enlarged. Its rhythm was normal and murmurs were not heard. Cardiac catheterization disclosed a marked pulmonary hypertension (32/45 mm. Hg). Radiological examinations of the chest revealed "a mass" in the posterior middle third of the mediastinum. Symptomatically, there had been little or no change in the patient's condition at the time of admission to this hospital in March of 1952.

On examination here the patient was slightly orthopneic, but there was no dyspnea at rest. Cyanosis and "clubbing" were not present and there was no venous engorgement or edema. The lungs were clear to percussion and auscultation. The heart had a normal rhythm and its sounds were loud and clear. Except for a short systolic blow, there were no cardiac murmurs. P2 was loud and split. Routine tests for syphilis and tuberculosis were negative. There was no anemia or leucocytosis. The sedimentation rate was 21 mm/hr. Radiological examinations of the chest revealed enlargement of the right side of

the heart; the left auricle was definitely not enlarged. The presence of an ill-defined posterior mediastinal mass was confirmed and the left main stem bronchus appeared narrowed. As had been noted previously, the lung fields showed vascular engorgement, and, in addition, there was a diffuse opacification of the right lower lung. Electrocardiograms were interpreted as showing right ventricular strain. A definite diagnosis could not be made at this time, although it seemed worthwhile to try irradiation therapy before attempting surgical exploration of the mediastinal mass. X-ray therapy was therefore started and the patient discharged. Although she had no further hemoptyses her exercise tolerance decreased and she experienced occasional fevers of 100–101°. The mediastinal mass remained unchanged. She was re-admitted on October 18, 1952. Again, numerous studies either confirmed previous findings or were diagnostically of no help. The only new development was the appearance of an infiltrative process at the left base in addition to that already described at the base of the right lung. She was again digitalized, but received little or no benefit after a two-week trial. Subsequently, on November 11, 1952, Dr. Alfred Blalock performed a left thoracotomy. The pulmonary artery was dilated and its pressure was obviously high. The left lung pulsated vigorously. The left auricle was entirely normal. On exploring the hilus of the left lung there was a stony mass (found on frozen section to be dense collagen) completely encasing the major bronchi, pulmonary veins, and part of the trachea. Excision of this material was impossible. Post-operatively, the patient did well for several days. On the ninth post-operative day she developed cyanosis, her respiratory rate suddenly increased and on the tenth day her blood pressure fell and she became cyanotic. Death occurred eleven days after the operation.

At *autopsy* (#24001) there was a recent left thoracotomy incision and the pleural space on this side contained a liter of blood-tinged fluid. The left pleural and pericardial surfaces were covered with fibrin. At the bifurcation of the trachea there was an ill-defined mass of dense, yellowish-white tissue which measured roughly 6 x 6 x 4 cm. Multiple slices revealed pigmented areas suggesting the remains of lymph nodes, and, in addition, there were white chalky foci apparently containing amorphous calcium. This fibrous mass completely encased both right pulmonary veins and narrowed their lumina to mere slits; they could be probed only with difficulty. It should be emphasized that although the left pulmonary veins were embedded in the fibrous mass, as noted at operation, they were not narrowed. The bronchi on both sides were also surrounded and slightly narrowed by this process. The main pulmonary artery was dilated and its left branch was enveloped but not appreciably narrowed by the fibrous mass. Histologically, this peculiar mass was composed of unusually thick, collagen fibers. It was relatively acellular and formed bundles running haphazardly in all directions, often forming whorls and concentric rings about small blood vessels. It had surrounded nerves, larger arteries, lymph nodes and bronchi, frequently becoming continuous with their connective tissue coatings. Most important of all for the present consideration was the fact, described in the gross, that it had completely encircled and markedly narrowed both pulmonary veins on the right side. Furthermore, intimal proliferation and organization of thrombi in these veins had left only tiny recanalized channels for the return of blood to the left auricle. It was clear microscopically that the apparently caseous areas seen grossly were nothing more than amorphous foci of disintegrating collagen, perhaps the result of infarction. There was nothing in many sections, including numerous special stains, to suggest an etiology for this collagenous mass. We have seen several cases (Case #5 is one) in which there have been similar collagenous masses, usually mediastinal, for which we have been unable to find a cause; a number have also been described in the literature (1) (3) (4) (5) (6), but obstruction of the pulmonary veins was mentioned in only one instance (1). The lungs were firm and heavy. The cut surface of the right lung, especially the lower lobe, presented a diffuse, gray induration which was not at all unlike the appearance of the lower lobes in Case #1. Scattered throughout a fine network of gray scar tissue were many grossly visible alveoli. In contrast, the left lung was red and congested, and, if anything, more edematous than the right, but there was no consolidation, induration or scarring. Although the right ventricle was definitely dilated and hypertrophied, the heart weighed only 320 grams. Microscopic sections of the heart revealed hypertrophy of the right ventricular fibers and the post-operative fibrinous pericarditis. There was nothing to suggest rheumatic fever. The liver was congested, grossly and microscopically. There was nothing of significance in the other organs.

Microscopic examination of many sections from the right lung, the side on which the veins were obstructed, revealed varying degrees of a severe and widespread, although focal interstitial fibrosis

(Fig. 3). With slight variation it was entirely similar to the process described in the previous cases, especially the first. The alveolar septa were everywhere greatly thickened by mature connective tissue. In some areas a few cells, mostly fibroblasts, were still present and in occasional areas proliferating small blood vessels were conspicuous. Foci of emphysematous alveoli were common. Practically all of the alveoli in the areas of fibrosis were lined by a low cuboidal epithelium. An occasional alveolus contained a few hemosiderin-laden macrophages. The arteries and arterioles in this lung, with obstructed veins, often had greatly thickened walls and small or even tiny lumina. In occasional arterioles and smaller arteries this was clearly due to medial hypertrophy and intimal proliferation. On the other hand, the larger arteries presented a somewhat different picture. Frequently, they, too, appeared to have chiefly intimal proliferation, though in some instances newly formed blood vessels, sometimes even with a muscular wall were embedded in the connective tissue in the original lumen of the vessel, such as one sees at times as the end result of an organized and recanalized thrombus. There was not only a complete absence of vascular changes in the left lung, the side with widely patent veins, there was also *no interstitial fibrosis*. Numerous sections of the left lung showed nothing more than the ordinary changes of chronic passive congestion.

Comment: In this case the right pulmonary veins were markedly obstructed as a result of compression by mediastinal collagenosis, and there was widespread interstitial fibrosis of the right lung. The left pulmonary veins were not obstructed and there was no fibrosis of the left lung.

Case #4

(Hist. No. A-73763). This 3 year old girl was admitted to the Johns Hopkins Hospital for the 6th and final time on December 23, 1952. Shortly after birth in November of 1949, she was found to have congenital cyanotic heart disease. The underlying nature of this condition was not suggested until March of 1950. At that time, on the basis of physical examination and fluoroscopy, tricuspid atresia with transposition of the great vessels was first offered as the most logical diagnosis; although right axis deviation in the electrocardiogram was somewhat puzzling. When she was seven weeks old she had an attack of "pneumonia" which was diagnosed and treated at another hospital. Digitalis was started at that time and continued for the rest of her life. She was admitted to the hospital twice in 1950, for study and possible cardiac surgery. The latter was, for various reasons, postponed on both occasions. In May of 1951, she was admitted for the third time because of digitalis intoxication. Her fourth admission in February of 1952 was for cardiac catheterization. These studies were not conclusive, but did tend to confirm the previous diagnosis. She was admitted for the fifth time on December 4, 1952, for angiocardiography. These studies confirmed the diagnosis of tricuspid atresia, although they were inadequate to determine whether or not there was an associated transposition of the great vessels. It should be stated that this child had always been cyanotic and that she did have some exertional dyspnea, although spells of paroxysmal dyspnea or loss of consciousness were never noted. Weight gain and development were slow. Except for occasional upper respiratory infections and the "pneumonia" at 7 weeks of age mentioned above, there had never been any evidence of parenchymal lung disease, either by x-ray or on physical examination. Following the admission for angiocardiography she returned home, but subsequently became extremely lethargic with transient periods of unresponsiveness. She finally became semicomatose and was admitted for the 6th and last time on December 22, 1952.

On physical examination she was noted to be undernourished, extremely cyanotic and responding only to painful stimuli. There was little or no "clubbing". The cardiac signs, consisting of enlargement and a moderately harsh systolic murmur in the 3rd and 4th interspaces, had not changed appreciably from previous examinations. Supportive therapy, including 100 cc. of whole blood, was to no avail. She remained stuporous and died early on the morning following admission.

At *autopsy* (#24067) the clinical impression of tricuspid atresia and transposition of the great vessels was confirmed, and the aorta and pulmonary artery arose from a single ventricle. The heart was enlarged. This was due primarily to dilatation and hypertrophy of the right auricle and the single

ventricle. A patent foramen ovale provided the only outlet for blood entering the right auricle. The ductus arteriosus had a 1 mm. lumen. There was a fresh thrombus in the right auricular appendage. The mitral, aortic, and pulmonary valves were all delicate. The pulmonary veins were most remarkable. On the right they were widely patent and perfectly normal, while on the left the only vestige of pulmonary veins were two "fibrous cords", each measuring about 2 mm. in overall diameter. Grossly, they did not appear to have a lumen. Microscopic sections of the heart confirmed the fresh right auricular thrombus and also disclosed a few small myocardial necroses. Under the microscope, except for a few tiny endothelialized channels, the left main pulmonary veins were mere cords of abnormally arranged muscle and connective tissue. The vestigial size of the veins, the malformation of their walls and the absence of scarring in the tissues about them, suggested that the obstructing process was a congenital rather than an acquired one. The lungs were heavy, weighing 500 grams. The pleura over the right lung was smooth and delicate; over the left lung, posteriorly and laterally, it was dull and thickened. On section, the left lung was firm and yellowish-gray, while in contrast, the right was considerably softer, red, and congested. The left lung was obviously the seat of a widespread fibrotic process, which, except for gross emphysema in its lower lobe, was quite similar in appearance to that of the preceding cases. There were a few, fresh, wedge-shaped infarcts in each lung, thought to be the result of emboli from the fresh mural thrombus in the right auricular appendage. Grossly and microscopically, the liver was markedly congested. There was a massive, unexplained, fresh hemorrhage in the wall of the stomach. The brain was grossly swollen, with flattened convolutions, while microscopically there were many perivascular hemorrhages. Significant changes were not present in other organs.

Sections from the left lung, the side on which the veins were congenitally stenotic showed a widespread and severe form of focal interstitial fibrosis of the type described in the previous cases (Figs. 4, 6). In view of the severity of the fibrosis it is surprising that it was not demonstrated in x-rays of the chest. Perhaps it was masked by the emphysema. The connective tissue in the alveolar walls was usually hyaline and relatively acellular, although in some areas fibroblasts were still evident. In a few areas this interstitial tissue contained an occasional plasma cell, lymphocyte or macrophage. Emphysema, as was noted in the gross, was considerably more prominent than in the preceding cases. It was focal and was most marked in or near the areas where the fibrosis was most extensive. Epithelialization of alveoli, a prominent feature of the process, was also present in this case. A few alveoli contained hemosiderin-laden macrophages; most were empty. The vascular changes in the left lung were identical with those in the involved portions of lung in the previous cases. Medial hypertrophy and intimal proliferation had caused marked narrowing of practically all arterioles and smaller arteries. Again some of the larger arteries presented the same type of recanalization as that described in the previous cases. Sections from the right lung revealed only edema and congestion; there was no interstitial fibrosis and all of the vessels appeared quite normal. The fresh infarcts noted grossly were also seen in the sections of both lungs and their age was compatible microscopically with that of the thrombus in the auricular appendage.

Comment: In this case, widespread interstitial fibrosis of the left lung was associated with extreme congenital stenosis of the left pulmonary veins. The pulmonary veins on the right side were normal and there was no fibrosis in the right lung.

Case # 5

(Hist. No. 664434). This 26 year old white male truck driver was admitted to this hospital for the first and only time on February 12, 1954, for diagnosis and treatment of "heart failure" of unknown etiology.

The first indication of any ill-health was rejection by the Merchant Marine, at age 17, on the basis of a heart murmur. Sometime after this the patient developed mild exertional dyspnea, paroxysm of cough and occasional hemoptyses. When he was 20, he described having had a 3 month period of almost daily hemoptyses, frequently raising a cupful of blood. He was finally hospitalized for this and with oxygen and digitalis therapy was sufficiently improved to be discharged at the end of a week.

Following discharge, he continued to have dyspnea and even developed some orthopnea, but was able to continue working. Two years later, when he was 22, severe diarrhea for one week necessitated hospitalization for the second time. He remained in the hospital for 6 weeks and was again digitalized. For the next three and a half years dyspnea continued, but otherwise there was little or no change in his condition until about 6 months prior to admission when dizziness, nausea and vomiting appeared. Six weeks before admission he became, rather abruptly, so orthopneic and so dyspneic that he was unable to move about and even eating exhausted him. He was forced to stop working. Four weeks before admission he developed, for the first time, abdominal swelling, ankle edema and slight cyanosis. His condition did not improve, and he was subsequently brought to this hospital and admitted on February 12, 1954.

At the time of admission his vital signs, except for a slightly elevated pulse, were normal. There was pitting edema of the ankles and feet. The lips, ears and nailbeds were mildly cyanotic, although there was no "clubbing". The neck veins were full. The chest was resonant to percussion. Medium moist inspiratory rales were heard posteriorly and laterally over the left chest and at the right base. The heart was enlarged both to the right and to the left. The precordium was active and there were systolic and diastolic impacts. A systolic thrill was also felt at the apex. Auscultatory findings were not entirely consistent among the various observers, although most agreed that the sounds were loud and clear, that there was a high-pitched apical systolic murmur, and that the pulmonic second sound was greater than the aortic second sound. The abdomen was distended and the liver palpable 7 cm. below the costal margin. He was not anemic. The sedimentation rate was 30 mm./hr. X-rays of the chest showed a markedly enlarged heart with a prominent pulmonary conus, and the lung fields were interpreted as showing increased broncho-vascular markings. Fluid was seen at the base of each lung, and in the interlobar fissures. Electrocardiograms showed right ventricular strain and digitalis effect. It was felt on admission, in spite of several inconsistencies, that the patient most likely had rheumatic heart disease with mitral insufficiency, and that evidence of mitral stenosis would become apparent as the heart became compensated. However, the latter was never achieved, and in spite of vigorous therapy his cyanosis deepened and his respiratory distress became even greater. A pericardial tap produced 150 cc. of pinkish fluid. His deterioration continued, and he died on the fourth hospital day without any satisfactory diagnosis having been made.

At *autopsy* (#24881), except for 200 cc. of clear, loculated fluid on the right side, both pleural spaces were practically obliterated by dense fibrous adhesions. The pericardial sac contained 200 cc. of blood-tinged fluid. At the bifurcation of the trachea there was an ill-defined mass of dense, yellowish-white tissue which measured roughly 4 x 3 x 2.3 cm. This tissue encased and markedly constricted all of the pulmonary veins; it did not involve the pulmonary arteries. At the narrowest point the lumina of the major pulmonary veins measured as follows: the right superior vein 2 mm.; the right inferior vein had a 4 mm. lumen; the left inferior vein measured 2 mm.; while the left superior vein, the most constricted of all, had only a tiny slit-like lumen. Multiple slices through the mass revealed several chalky areas, quite like those seen in Case #3. The remains of an occasional pigmented lymph node were also found embedded in this fibrous tissue. At its periphery the mass was often continuous with the scar tissue which covered large areas of both lungs. However, at no point did it appear to invade the lung parenchyma. It is sufficient to say that microscopically this collagenous mass was identical with the one described in Case #3. The lungs were very heavy and together weighed 3020 grams. The bronchi contained pink edema fluid, but were not narrowed by the mediastinal mass. The pulmonary arteries were dilated and thick walled. On section, both lungs were markedly congested and edematous. In addition, both lower lobes were indurated and seemed to contain excessive amounts of connective tissue. After fixation, scattered small foci of a yellow or rusty pigment became apparent. The heart, weighing 450 grams, was enlarged by tremendous dilatation and hypertrophy of both the right auricle and the right ventricle. The wall of the latter measured 8 mm. in thickness, while the left ventricular myocardium measured 11 mm. There were no valvular or endocardial lesions. Except for congestion of the liver and enlargement of the spleen there were no other significant abnormalities.

Microscopic examination of many sections from all lobes of the lungs in this case revealed the most extreme changes of chronic passive congestion. Alveolar capillaries were everywhere greatly dilated and frequently bulged into the alveolar spaces. Some alveoli were filled with edema coagulum and "heart failure" cells. Iron-encrusted fibers, ("Gamna-Gandy bodies",) were common in the lower

lobes (Fig. 7) and were often found within multinucleated giant cells. Rounded, bluish-black structures resembling the Schaumann bodies of sarcoid were present both within giant cells and free in the tissues (Fig. 7). The focal interstitial fibrosis in most places was not as severe or as fully developed as in the first, third and fourth cases. However, in many sections from all lobes, especially those from the left upper lobe (Fig. 5), there was extensive proliferation of fibroblasts within many alveolar walls, and furthermore in some areas the process was even becoming relatively acellular and mature. Although the process was entirely similar to that seen in the preceding cases, there was no great thickening of alveolar walls with distortion of alveolar spaces and in only a few areas was there alveolar epithelialization.

Comment: In this case there was interstitial fibrosis of all lobes associated with marked narrowing of all the main pulmonary veins. The latter had been constricted by mediastinal collagenosis of the same type as that in Case #3. It is of interest, although not surprising, that there was more interstitial fibrosis in the left upper lobe than in any of the others and that this was the lobe with the most markedly constricted vein.

DISCUSSION

Five cases are described which demonstrate that obstruction to the outflow of a pulmonary vein can cause a severe and in some respects an unusual form of interstitial fibrosis in the lung parenchyma which it drains. The venous obstruction that resulted in interstitial fibrosis in these cases was produced by a variety of causes. In Case #1 the free flow of blood from the pulmonary veins into the left auricle was obstructed by a large myxoma; in Case #2 a similar obstruction resulted from a large auricular thrombus; in Cases #3 and #5 pulmonary veins were constricted by the dense collagen of mediastinal collagenosis; and in Case #4 there was congenital stenosis of pulmonary veins.

In order that the unusual nature of this interstitial fibrosis be clearly understood it must be emphasized that in its most fully developed form the interstitial fibrosis resulting from pulmonary vein obstruction is in no way different from certain others, especially the late stages of the "acute diffuse interstitial fibrosis" described by Hamman and Rich (7). It is only in its earlier developmental stages that this process appears unusual. This fact was appreciated only in retrospect, and is more or less a composite picture formed by the study of numerous sections from all five cases. It would therefore seem appropriate to outline its development and also to indicate the reasons for its uniqueness. As one might suspect, the earliest or mildest changes in the parenchyma drained by a partially obstructed vein are simply those of passive congestion; i.e., edema and capillary engorgement. However, if the venous obstruction is severe enough the alveolar walls become swollen and the normal structures within them separated by edema fluid. Proliferating capillaries and scattered fibroblasts eventually appear in these edematous alveolar walls and a delicate network of collagen is formed, while concurrently the alveoli usually become lined by cuboidal epithelial cells (Fig. 6). The collagen fibers thus formed gradually

become thicker, more numerous and more compact, and as the process matures the capillaries tend to disappear, so that in the final stages the alveolar walls are markedly thickened by a relatively acellular and avascular connective tissue (Figs. 1, 3, 4.). Although a few macrophages, lymphocytes or plasma cells may be seen within the alveolar walls during the development of this process, it does not show as marked an infiltration of inflammatory cells as one sees in the early stages of the "acute diffuse interstitial fibrosis" of Hamman and Rich, although again it should be pointed out that in the final fibrous stage the alveolar walls in the two conditions are quite indistinguishable. Furthermore, in the present cases, in contrast to diffuse interstitial fibrosis, there are no hyaline membranes or large plaque-like cells lining the alveoli, and although alveolar epithelization is a prominent feature, there is no metaplastic proliferation of bronchiolar epithelium such as occurs in diffuse interstitial fibrosis. Foci of hemosiderin-laden macrophages are scattered in the alveoli, and in one case (Case #5) there were iron encrustations ("Gamna-Gandy bodies") and also dark, hematoxylin-staining bodies, some of which resembled Schaumann bodies (Fig. 7). Many of the arterioles and smaller arteries in the *involved areas* become markedly narrowed by medial hypertrophy and intimal proliferation, while the arteries and arterioles in uninvolved areas of lung remain quite normal. The narrowing of some of these vessels in the involved areas is also clearly the result of thrombosis, organization and recanalization. Although these arterial and arteriolar thrombi are an important anatomical feature of this process, they do not play a primary or etiological role in its development since it is well known that pulmonary arterial and arteriolar thromboses do not produce interstitial fibrosis of the sort described in the present cases.

Since this peculiar form of pulmonary fibrosis results from extreme pulmonary venous hypertension, it may be remarked that similar *focal* areas of fibrosis occur in rare instances of very severe and protracted pulmonary venous hypertension resulting from cardiac failure, particularly mitral stenosis. Why, in those rare instances the process is focal, rather than general throughout the lung, has never been clear and the explanation of this must await future studies. The answer to this problem may possibly reside in circulatory factors, such as arterio-venous shunts, that govern the flow of blood through different parts of the lung. In this connection it is pertinent that, as shown in the present cases, marked obstruction of a main pulmonary vein does not produce interstitial fibrosis uniformly throughout the lobe drained by that vein. Even though the fibrosis may be severe and widespread, even with fairly large confluent areas, it is still focal with intervening areas of normal lung.

Edwards and Burchell (1) have reported a case in which the veins from four of the pulmonary lobes were markedly narrowed by a "fibrosing mediastinitis," while the vein from the fifth lobe was spared. This study was concerned with pulmonary arterial alterations that occur in cases of pulmonary venous hyper-

tension, rather than with alterations in the pulmonary parenchyma. In one sentence it is mentioned that in the lobes drained by the obstructed pulmonary veins "... there were foci in which the alveolar walls were thickened by collagen", and also that in these areas the alveoli were lined by simple cuboidal epithelium. It is probable that this fibrosis was of the type described in the present cases.

Although we have not had the opportunity of examining a case of so-called "idiopathic pulmonary hemosiderosis," a study of the cases reported in the literature (8, 9) would indicate that the anatomical changes in the lungs in this condition are very similar to the changes produced by pulmonary vein obstruction. In view of this similarity, and the obscureness of the etiology of the former, and even in spite of certain differences, such as the characteristic anemia and the diffuseness and symmetry of the process within the lungs in idiopathic pulmonary hemosiderosis, it would be worthwhile to carefully examine the pulmonary veins in all cases of idiopathic pulmonary hemosiderosis, although it may well be that the pulmonary fibrosis in this peculiar condition has a different pathogenesis.

The effect on the lungs of ligation of one or more pulmonary veins has been studied experimentally (10) (11) (12) (13) (14) (15). In addition to hemorrhage, infarction and infection, "focal scars" have been described in some instances, but no interstitial fibrosis comparable to that in the present cases has been found in the literature. Furthermore, with one exception, these experiments all consisted of sudden and complete ligation of one or more pulmonary veins, and could therefore, probably be expected to have results quite different from those of gradual and partial stenosis. Cheng (13) has reported partial and complete ligations of the pulmonary veins in rats. Both procedures led to hemorrhage, infarction and focal scarring, but there were no lesions similar to those in the present cases. Since the results of his partial ligations were the same as those of his complete ligations, and since no examination of the patency of the veins at autopsy is mentioned, one cannot be sure whether the veins were partially or completely obstructed, particularly since the ligatures were loosely placed at operation and then tightened extra-thoracically three or four days later. At any rate, it would be worthwhile to study carefully the effects of partial pulmonary vein obstruction in a larger animal in which the degree of obstruction can be determined more readily than in the rat.

It is difficult, if not impossible, from the material at hand to outline a characteristic clinical picture for this pathological entity. In the first and second cases the clinical picture was dominated by the underlying disease process, which secondarily led to the obstruction of the pulmonary veins. In the fourth case the clinical picture was primarily that resulting from the cardiac malformation. We are left then with only the third and fifth cases which, at least in retrospect, manifested themselves chiefly by the results of pulmonary vein

obstruction. In these two cases there was dyspnea, cough and repeated hemoptyses. Hemoptysis and dyspnea also occurred in Case #2. There is no record of hemoptysis in the history of Case #1, and Case #4 was a child at an age in which the production of sputum is uncommon. Cases #3 and #5 would then suggest that pulmonary vein obstruction be considered in any obscure illness with dyspnea, cough and repeated hemoptyses, particularly if there is an enlarged right heart, and also if the duration of these symptoms would tend to rule out carcinoma of the lung and if mitral stenosis or severe left heart failure appears unlikely. In view of the severity of the lesions at autopsy in the present cases, it is surprising that x-rays of the lungs revealed evidence of parenchymal lung disease other than nonspecific signs of congestion, only in cases #1 and #3. However, in these two cases there were *persistent* areas of opacification, which at the time were puzzling. The reasons for this incomplete correlation between the x-rays of the lungs and the severity of the lesions at autopsy are not entirely clear at the present time and must await further study.

SUMMARY

The clinical course and autopsy findings of five cases are presented, which demonstrate that obstruction of a pulmonary vein from any cause leads to an unusual form of interstitial fibrosis in the lung parenchyma drained by the obstructed vein. These cases would suggest that in any instance of unexplained pulmonary interstitial fibrosis the pulmonary veins and their auricular orifices should be carefully examined. The possibility of recognizing this condition before death is briefly discussed.

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EXPLANATION OF ILLUSTRATIONS

(Photographs by Miss Patricia E. Whitney)

Fig. 1. Case #1. Typical area of late stage of interstitial fibrosis.

Fig. 2. Case #2. Interstitial fibrosis. Hemosiderin-laden macophages in alveoli.

Fig. 3. Case #3. Interstitial fibrosis with prominent alveolar epithelialization and scattered hemosiderin-laden macrophages.

Fig. 4. Case #4. Similar to Fig. 3, Case #3.

Fig. 5. Case #5. Interstitial fibrosis.

Fig. 6. Case #3. High power view of early stage of interstitial fibrosis with edematous alveolar walls, proliferating capillaries, fibroblasts and occasional round cells. Also shows alveolar epithelialization.

Fig. 7. High power view showing iron encrusted fibers (A), and masses in multinucleated giant cells resembling Schaumann bodies (B).

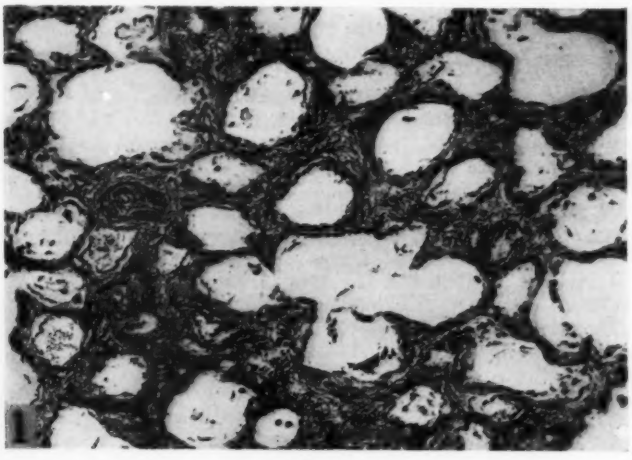
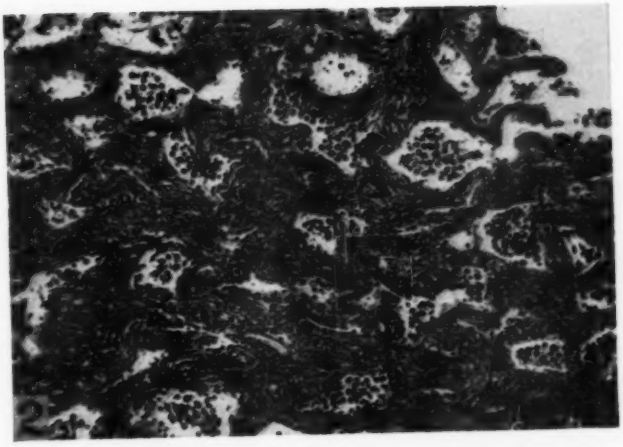
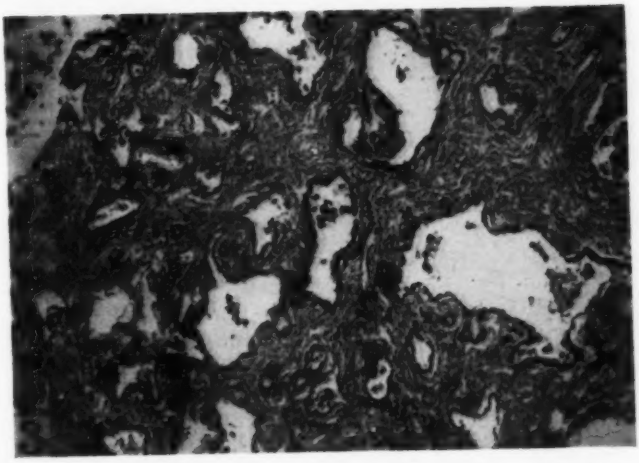
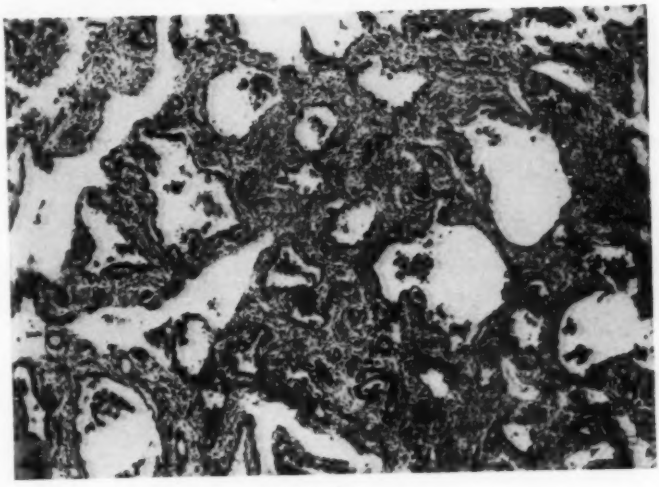


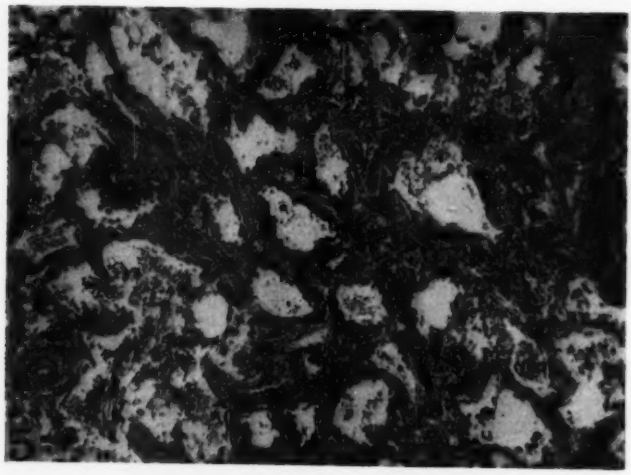
Fig. 1

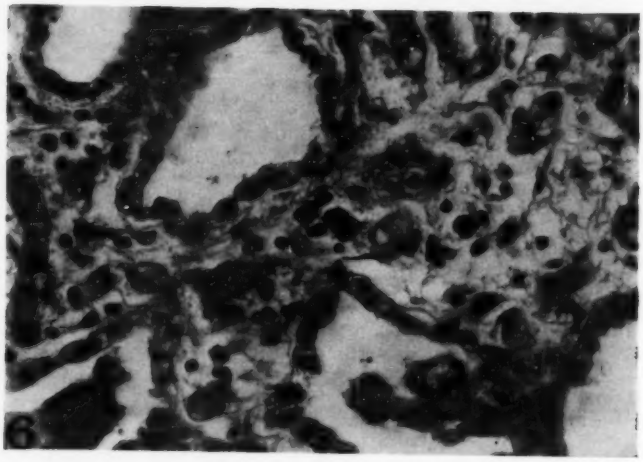


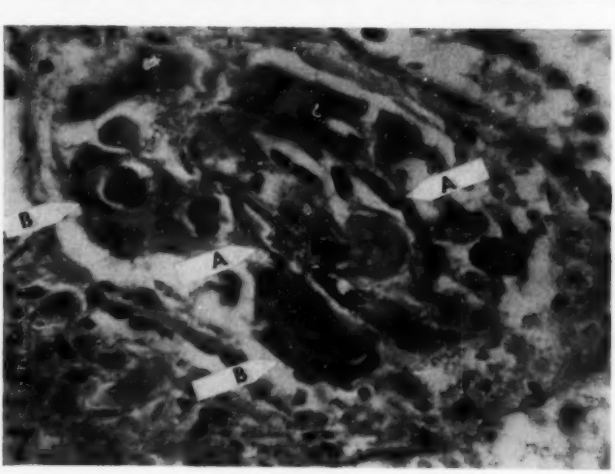




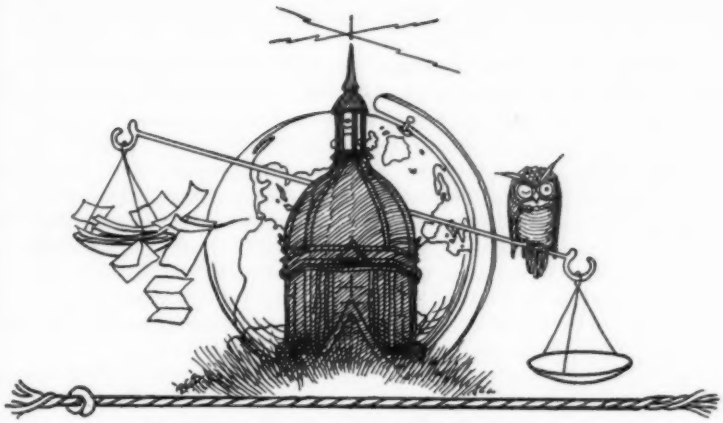
FIGS. 2–4







FIGS. 5–7



BOOK REVIEWS

The Nature of Brucellosis. By WESLEY W. SPINK. 464 pp. $8.00. The University of Minnesota Press, Minneapolis, Minnesota.

Dr. Spink is recognized the world over as an authority on brucellosis, and this book gives added support to such recognition. An introduction containing a detailed and interesting history of brucellosis is followed in succeeding chapters by discussions of the biological characteristics of the etiologic organism and the epidemiology of the disease in animals and its reflection in the patterns of human infection. A section on pathogenesis presents concepts of interest not only for the infection under discussion, but also sheds light on possible mechanisms by which other intracellular parasites produce disease. The discussion of brucellosis in relation to other "granulomatous" diseases and the conclusions Dr. Spink suggests in terms of therapeutic principles are significant and provocative.

Detailed descriptions of the clinical aspects of brucellosis are enhanced by an appendix containing summaries of 244 cases seen in the University of Minnesota clinics. Arrangement in this fashion permits the reader to refer to specific cases cited in support of various points in the text.

It is difficult to find fault with this excellent monograph on the nature of a disease, and it can be recommended without reservation to all who are interested in the biology of infection, be they microbiologists, pathologists or clinicians.

ROBERT W. TREVER

Roentgen Signs in Clinical Diagnosis. By ISADORE MESCHAN with the assistance of R. M. F. FARRER-MESCHAN. 1058 pp., $20.00. W. B. Saunders Co., Philadelphia, Pa.

This book intends to cover the entire field of diagnostic roentgenology. The chapters are arranged according to groups of similar roentgen signs, clinical entities, or anatomic systems. The fundamental background for radiographic technic and protection from roentgen irradiation is given. A large number of diagrammatic sketches illustrate the positioning of the patients for the various standard roentgenographic procedures. Most of these illustrations are taken from the author's previous book *An Atlas of Normal Radiographic Anatomy* published by Saunders in 1951. These diagrammatic sketches will undoubtedly be of great help to the general practitioner attempting to do his own radiography.

A short clinical outline, the pertinent laboratory findings, and a short clinical pathological correlation are given for most disease entities: these enable a better understanding

of the radiographic findings. It is noted, however, that the terminology used is occasionally vague and that certain clinical and pathological information is not up to date. The differential diagnoses for roentgenologic findings are frequently incomplete. At the end of each chapter, there is a bibliography referring to both classical and current articles on each subject. Probably the greatest shortcoming of this text is the frequent use of cross references to an entire chapter. Since the material is grouped according to roentgen signs, a specific disease entity is often described in several chapters. The system of cross indexing and cross reference does not allow the reader to compile all the roentgen manifestations of a certain disease without considerable difficulty. One of the most outstanding features of this text is the frequent use of diagrams to explain the accompanying radiographs. This will facilitate the study of such a text for the medical student and beginner in radiology. The reproductions of the roentgenograms are excellent.

The book will be of greatest value to the general practitioner doing a limited amount of roentgenologic work, the medical student, and the beginner in radiology. It can be used both as a quick reference for roentgenographic positioning and for differential diagnosis of roentgenologic signs.

ERICH K. LANG

Management of Emotional Problems in Medical Practice. Edited by SAMUEL LIEBMAN. 152 pp., $5.00. J. B. Lippincott Co., Philadelphia, Pa.

This book is the publication of a series of nine papers given by guest lecturers of no little repute at the North Shore Health Resort, Winnetka, Illinois. Thus, there is a chapter by Karl M. Bowman on psychiatric emergencies including suicide attempts, threats, mass panic, etc., an essay by Francis J. Gerty on sedatives and stimulants that is not only lucid and concise but of an enviable literary style, a chapter by Franklin G. Ebaugh devoted to the depressed patient, a contribution by Leo H. Bartemeier on management of overeating, overdrinking, and oversmoking, a chapter by Walter C. Alvarez anecdotally describing how patients can be made apprehensive by the misinterpreted or inept comments of the physician, a chapter by Lewis L. Robbins on anxiety, a chapter by Marc H. Hollender on the utilization of community resources in medical practice, etc. Chapters by George C. Ham on the management of the multiple complainer and by Otto Billig on the management of emotional reactions in the male involutional period are especially well done, clinically illustrated, versions of the comprehensive approach to the somatically ill patient. They imply that the understanding of the meaning of patient's the symptoms in the broadly personal sense is as crucial as the diagnosis and treatment of specific disease processes. Dr. Ham stresses the importance of making a "total diagnosis" and lists a number of guides to the psychologically sound management of patients with multiple somatic complaints.

This publication is aimed at all physicians with clinical practices, and not the psychiatric specialist, particularly. It is not, as one could mistakenly be led to expect from the title, a comprehensive and systematic treatment of clinical management of emotional problems; but the range of topics included is sufficiently broad, and each chapter is self-contained and (with minor exceptions) intelligibly and concisely written, so it is eminently suitable for browsing for topics of special interest to each reader.

FREDERICK J. ZIEGLER

Proctology. By HARRY E. BACON, STUART T. ROSS AND PORFIRIO MAYO RECIO. 441 pp., $10.00. J. B. Lippincott Co., Philadelphia, Pa.

This volume succeeds admirably in following out the purposes of the authors in "presenting the fundamentals of basic proctology shorn of historical data, extensive description of pathology, hypotheses of etiology, obsolete forms of examination and treatment, and

bibliography." It is a good textbook of 441 pages, valuable for student or practitioner, and covering proctology in its broadest definition, namely, the science that treats of surgical diseases of the colon, rectum, and anal canal. It is an outgrowth of the original *Essentials of Proctology* published in 1943 by the senior author. The descriptive matter is well arranged and clearly written. Starting with the essentials of anatomy and physiology of the regions involved, it concludes with a chapter on parasitic diseases of the lower intestinal tract. The text is abundantly and appropriately illustrated by drawings, roentgenograms, and photographs, some of the latter in color. Many of these illustrations have been made available through other publications and texts of the authors, notably, *The Atlas of Operative Technic* (Bacon and Ross). Finally, the test bears the stamp of authority, because the senior author of all contemporary men has probably done more to write and teach about this subject, and, indeed, to make proctology emerge as the impressive surgical specialty that it has become in this country today. Although his procedures have at times been the subject of controversy (as in the "pull through" operation for cancer in the upper and mid-rectum) he stands as an experienced observer, dynamic teacher, and active practitioner in this field.

SAMUEL MCLANAHAN

A New Approach to Schizophrenia. By JULIUS I. STEINFELD. 195 pp., $4.95. Merlin Press, Inc., New York, N. Y.

These posthumously published essays and case histories are variations on the theme that the core of the schizophrenic process is a disturbance of the vegetative nervous system, produced by a "vegetative trauma" in earliest infancy. The trauma is near-starvation with resulting disturbance of the acid base balance and overstimulation of hypothalamic centers. It is well recognized that adult schizophrenics often give evidence of dysfunctioning of the autonomic nervous system, and the study of the nature of this dysfunction has become a promising field of research. That it may be related to early feeding difficulties is an interesting hypothesis which, however, is hard to verify because data on earliest infancy are difficult to obtain. Dr. Steinfeld illustrates it with forty concise, well presented case histories of schizophrenics who seem to have had severe feeding difficulties in infancy.

The hypothesis is in accord with the frequency of gastro-intestinal complaints and eating problems in schizophrenics, and with the difficulties in attempting to cure them by purely verbal means, which the author relates to the fact that the initial trauma occurred before the infant could speak. It is also consistent with other current views concerning the nature of the mother-child relationship in schizophrenics.

Unfortunately, the neurophysiological aspects of the hypothesis are so loosely stated that they cannot be checked. On the psychological side the author, as so often happens, is carried away by his idea and tries to stretch it to cover all sorts of psychopathological manifestations in schizophrenics including homosexual behavior, catatonia, and sado-masochism. The speculations are often so far-fetched and the alleged supporting evidence so tenuous that they detract from rather than add to the plausibility o his central thesis. In this book as in his previous one, however, Dr. Steinfeld gains the reader's respect as an astute, resourceful, compassionate psychiatrist who showed remarkable imagination and ingenuity in trying to help his patients.

JEROME D. FRANK

Essays in Biochemistry. Edited by SAMUEL GRAFF. 345 pp., $6.50. John Wiley & Sons, Inc., New York, N. Y.

This collection of twenty-five essays in biochemistry was written in honor of Professor Hans Thacher Clarke on the occasion of his retirement as Professor and Chairman of the

Department of Biochemistry, College of Physicians and Surgeons, Columbia University. The authors were all former students or academic associates of Professor Clarke. Although volumes of scientific papers by students and associates have often been collected in the past to honor the retirement of a distinguished professor, it is not often that such a volume exceeds the bounds of local interest, as this one does, and becomes a widely important and authoritative statement of contemporary work and thought.

According to the preface the essayists were accorded free rein in regard to subject and style. Some are critical reviews of the current status of certain biochemical problems; others are speculative and philosophical in outlook. The wide range of interests of Professor Clarke's associates and students may be indicated by a few titles: "Some Metabolic Products of Basidiomycetes", "The Biochemistry of Lysogeny", "The Chemical Basis of Heredity Determinants", "Tetrazoles as Carboxylic Acid Analogues", "On the Nature of Cancer", and "The Biosynthesis of Porphyrins". The imprints of both personality and scientific outlook are clearly seen in articles with such provocative titles as "The Very Big and the Very Small: Remarks on Conjugated Proteins", "Unbalanced Growth and Death: A Study in Thymine Metabolism", and "On the Bigness of Enzymes".

The book can be highly recommended to students of biochemistry as an interesting and provocative tour through many phases of biochemistry and neighboring sciences. In addition it is a remarkable tribute to the stewardship of Professor Clarke that such an outstanding group of original investigators, as represented by the authors of this volume, should have developed their scientific interest and outlook in their association with him.

ALBERT L. LEHNINGER

BOOKS RECEIVED FOR REVIEW

Clinical Pathology. Application and Interpretation, 2nd edition. By BENJAMIN B. WELLS. 488 pp., $8.50. W. B. Saunders Co., Philadelphia, Pa.

Diabetes Mellitus. Handbook for Physicians. By HOWARD F. ROOT AND PRISCILLA WHITE. 346 pp., $7.00. Landsberger Medical Books, Inc. Distributed solely by The Blakiston Division of the McGraw-Hill Book Co., New York, N. Y.

Dictionary of Poisons. By IBERT AND ELEANOR MELLAN. 150 pp., $4.75. Philosophical Library, New York, N. Y.

Diseases of the Breast. By C. D. HAAGENSEN. 751 pp., $16.00. W. B. Saunders Co., Philadelphia, Pa.

Diseases of the Heart, 2nd edition. By CHARLES K. FRIEDBERG. 1161 pp., $18.00. W. B. Saunders Co., Philadelphia, Pa.

Heritable Disorders of Connective Tissue. By VICTOR MCKUSICK. 224 pp., $7.50. The C. V. Mosby Co., St. Louis, Missouri.

Pediatrics. Edited by DONALD PATERSON AND JOHN FERGUSON MCCREARY. 654 pp., $14.00. J. B. Lippincott Co., Philadelphia, Pa.